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PROGRESS REPORT

on

GRAIN AND FORAGE CROPS RESEARCH

of the

UNITED STATES DEPARTMENT OF AGRICULTURE

This progress report includes a summary of the current research of the USDA on grain and forage crops and a preliminary report of progress made during the preceding year. It is primarily a tool for use of scientists and administrators in program coordination, development and evaluation; and for use of advisory committees in program review and development of recommendations for future research programs.

The summaries of progress on USDA and cooperative research include some tentative results that have not been tested sufficiently to justify general release. Such findings, when adequately confirmed, will be released promptly through established channels. Because of this, the report is not intended for publication and should not be referred to in literature citations. Copies are distributed only to members of Department staff, advisory committee members and others having a special interest in the development of public agricultural research programs.

This report also includes a list of publications reporting results of USDA and cooperative research issued between July 1, 1967, and June 30, 1968. Current agricultural research findings are also published in the monthly USDA publications, Agricultural Research and The Farm Index.

> RESEARCH PROGRAM DEVELOPMENT AND EVALUATION STAFF UNITED STATES DEPARTMENT OF AGRICULTURE

> > Washington, D.C.

December 31/1968

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- 1. Farm Resources & Facilities
- 2. Utilization
- 3. Human Nutrition & Consumer Use
- 4. Marketing
- 5. Agricultural Economics
- 6. Forestry
- 7. Animal & Animal Products

- 8 Cotton
- 9. Grain & Forage Crops
- 10. Horticultural Crops
- 11. Oilseed & Peanut Crops
- 12. Plant Science & Entomology
- 13. Sugar
- 14. Tobacco

The source materials used by the advisory committees include organizational unit progress reports and subject matter progress reports. The latter contain information which was first reported in the organizational reports and has been assembled for use by commodity committees. The number prefixes shown below refer to advisory committees listed above.

ORGANIZATIONAL UNIT PROGRESS REPORTS

Agricultural Research Service (ARS)

1 - Agricultural Engineering

- 1 Soil and Water Conservation
- 2 Utilization -- Eastern
- 2 Utilization -- Northern
- 2 Utilization -- Southern
- 2 Utilization -- Western
- 3 Human Nutrition
- 3 Consumer & Food Economics
- 4 Market Quality
- 4 Transportation & Facilities
- 7 Animal Husbandry
- 7 Animal Disease & Parasite
- 12 Crops
- 12 Entomology

Economic Research Service (ERS)

- 1, 5 Economic Development
- 4, 5 Marketing Economics
 - 5 Farm Production Economics
 - 5 Economic & Statistical Analysis
 - 5 Foreign Development & Trade
 - 5 Foreign Regional Analysis
 - 5 Natural Resource Economics
 - 6 Forest Service Research (FS)
- 4, 5 Farmer Cooperative Service (FCS)
- 4, 5 <u>Statistical Reporting Service (SRS)</u>

SUBJECT MATTER PROGRESS REPORTS

- 6 Forestry (other than Forest Service)
- 7 Animal-Poultry & Products Research other than Husbandry,
 Disease and Parasite
- 8 Cotton and Cottonseed
- 9 Grain and Forage Crops
- 10 Horticultural Crops
- 11 Oilseed and Peanut Crops
- 13 Sugar
- 14 Tobacco

A copy of any of the reports may be requested from W. C. Dachtler, Executive Secretary, Grain and Forage Crops Research Advisory Committee, Research Program Development and Evaluation Staff, U.S. Department of Agriculture, Washington, D.C. 20250.

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INTRODUCTION

This report on grain and forage crops research covers work directly related to the production, processing, distribution and consumption of grain, rice, feed, forage and seed and their products. It does not include extensive cross-commodity work, much of which is basic in character, which contributes to the solution of not only grain and forage crop problems but also to the problems of other commodities. Progress on cross-commodity work is found in the organizations' unit reports of the several divisions.

The report is organized by "Problem Areas" which are shown in the table of contents. For each area there is (1) a tabulation of the number and location of the scientist man-years involved in research by USDA, (2) a statement of the research problem and objectives, (3) a summary of progress during the past year, and (4) a list of publications resulting from USDA and cooperative work.

Research on grain and forage crop problems is supported by (1) Federal funds appropriated to the research agencies of the USDA, (2) Federal and State funds appropriated to the State agricultural experiment stations, and (3) private funds for research carried on in private laboratories or for support of State station and USDA work.

Research by USDA

Farm research in the Agricultural Research Service comprises investigations on introduction, breeding and genetics, variety evaluation, culture, diseases, nematodes and weed control, insects, and crop handling and harvesting equipment and structures. This research is carried out in the Crops, Entomology, and Agricultural Engineering Research Divisions. It involves approximately 315 scientist man-years.

Nutrition, consumer and industrial use research in the Agricultural Research Service pertains to composition and nutritive value, physiological availability of nutrients and their effects, new and improved methods of preparation, preservation and care in homes, eating establishments and institutions, new and improved food, feed, and industrial products and the processes related to grains, rice, and forages. It is carried out in the following research divisions: Northern, Southern, and Western Utilization; Human Nutrition; and Consumer and Food Economics. The work involves approximately 175 scientist man-years

Marketing and economic research is carried out in four services. Grain, rice, feed, forage, and seed research in the Agricultural Research Service deals with physical and biological aspects of assembly, packaging, transporting and storing, and distribution. It is carried out by the Market Quality and Transportation and Facilities Research Divisions. Work in the Economic Research Service deals with marketing costs, margins, and efficiency; market potential; supply and demand; and outlook and situation. Consumer preference studies and research to improve crop estimates are carried out by the Statistical Reporting Service. Research on cooperative marketing is conducted by the Farmer Cooperative Service. The grain and forage research in these Services involves approximately 50 scientist man-years.

Interrelationships Among Department, State and Private Research

A large part of the Department's research is cooperative with State experiment stations. Many Department employees are located at State stations and use laboratories and office space close to or furnished by the State. Cooperative work is jointly planned, frequently with representatives of the producers or industry affected participating. The nature of cooperation varies with each study. It is developed so as to fully utilize the personnel and other resources of the cooperators, which frequently includes resources contributed by the interested producers or industry. There is regular exchange of information between station and Department scientists to assure that the programs compliment each other and to eliminate unnecessary duplication.

Privately supported grain and forage crops research emphasizes the solution of specific production, processing, and marketing problems. Much of it utilizes the results of more basic work done by State stations and Department scientists. For example, private research is devoted to the synthesis and evaluation of chemicals as herbicides, fungicides, and insecticides, the development of new combinations of materials for use as fertilizers, product and process development, improvements in equipment for planting, cultivating, harvesting, handling, processing, drying and storing grains, forages and seeds. Private research in marketing and economics is largely concerned with studies of consumer preferences, market potential, promotion and market development.

The contributions of producers of grain and forage crops and of related industries to the work of State stations and the Department have been an important factor in the success of public research programs. Producers, processors, and distributors offer land, products, and facilities for the testing of equipment and practices used in the production and distribution of grain, rice, feed, forage, and seed and their products.

I. FARM RESEARCH

BARLEY BENEEDING, GENETICS, DISEASES, QUALITY AND PHYSIOLOGY

Crops Research Division, ARS

USDA and Cooperative Program

	: Scientist Man-Years F.Y. 1968											
Location of Intramural Work		: Research Problem Area :										
	:	307	:	208	:	207	:	405	:			
	:		:		:		:		:			
Barley	:		:		•		:		:			
Maryland (Beltsville)	:	2.8	:	0.9	:	0.2	:	-	:	3.9		
Arizona	:	1.0	:	-	:	_	:	_	:	1.0		
California	:	1.0	:	-	:	_		-	:	1.0		
Idaho	:	0.2	•	-	:	-	:	-	:	0.2		
Michigan		0.8	:	_	:	0.5	:	_		1.3		
Montana	:	1.0	•	-	:	-		_	:	1.0		
North Carolina	:	_	:	1.0	:	_	:	-	:	1.0		
North Dakota		_	:	1.0	•	-	:	_	:	1.0		
South Dakota		0.6		-	:	1.0	:	_	:	1.6		
Wisconsin	:	_	:	-	:	_	•	4.8	:	4.8		
Total	:	7.4	:	2.9	:	1.7	:	4.8	:	16.8		

Intramural program is supplemented by extramural support representing (a) 0.9 SMY's at State Agricultural Experiment Stations $\underline{1}$, and (b) P.L. 480 funds in 4 countries representing 126,700 U.S. dollars equivalent.

^{1/} RPA 307, 0.7 and RPA 207, 0.2.

Problems and Objectives

The annual farm value of the barley crop amounts to \$500,000,000. Thirty percent of the crop is used by industry to manufacture malt and malt products and the remainder is used for feed and seed. It is estimated that future needs for barley will increase substantially for both feed and industrial use. To attain these goals, barleys with higher yield potentials must be developed by breeding. This can be accomplished by the use of hybrid barley, greater disease and insect resistance, stiffer straw, improved malting and feeding quality, and tolerance to stress conditions caused by acid or alkaline soils, drought, and winterkilling.

Major objectives of the research are to:

- 1. Develop hybrid barley for the major barley-growing areas.
- 2. Increase disease and insect resistance of barley varieties and hybrids.
- 3. Increase straw strength for greater use of fertilizer and improved cultural practices.
- 4. Breed varieties tolerant to soil and weather stresses.
- 5. Develop improved barleys for malting and feeding quality, and find better methods for evaluating quality.

Progress - USDA and Cooperative Program

-- Barley --

RPA 307 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY

A. Breeding and Genetics

1. Hybrid barley. During the year the first hybrid barley, Hembar, was grown in Arizona. Yield advantage of the hybrid ranged from 15 to 35 percent over the best adapted variety. Seed set on the male sterile female parent ranged from 30 to 80 percent of the maximum obtainable. The system for hybrid barley is based on genetic controlled male sterility rather than on the cytoplasmic one used for other crops. This genetic system (known as the "balanced tertiary trisomic system") makes use of an extra chromosome that can be genetically programmed to regulate several critical genetic events in steps leading up to the production of the male sterile female parent of the cross. The extra chromosome, however, is automatically eliminated in the last step of the method and thus does not participate in the crop grown by the farmer. The method can be used for other crops having a genetic makeup similar to barley. This includes about half of our cultivated crops.

- 2. New lines and germ plasm released. The female parent seed stock of Hembar (balanced tertiary trisomic line 63-j-18-17) was released jointly with the Arizona Agricultural Experiment Station. In addition, a bulk of balanced tertiary trisomic ($2\sqrt[4]{ms_2}$) lines with sessile lateral flowers was also released to plant breeders as a germ plasm source from which female parents for hybrids may be selected. A group of 216 genetic male sterile stocks including 19 known to be nonallelic and 33 not yet checked for allelism, were released jointly. These stocks are available in varying backgrounds in both spring and winter types, and are available to barley breeders for genetic and breeding material.
- 3. New varieties released. Four new varieties of barley were released. Steveland was developed in a cooperative program and released with the Idaho and Oregon Agricultural Experiment Stations. It is a spring feed barley which produces high yields under irrigation in southern Idaho and eastern Oregon. It has outyielded Trebi, Bonneville, and Gem by 7 to 9 bushels per acre in the recommended areas. It is early maturing, has strong straw, is superior to Bonneville, and equal to Trebi and Gem in threshability. It's test weight is higher than Bonneville and Gem, but slightly lower than that of Trebi.

Harland, a spring, feed barley, was developed in a cooperative program with the California Agricultural Experiment Station at Davis. It is a population variety as contrasted with a pureline variety and is predominantly 6-rowed with white covered kernels. The population will have a 2-week range in maturing, 1 to 2 percent male sterility and about 40 percent detectable heterozygosity. Harland is recommended from Fresno to Red Bluff, California and will equal or exceed the yield of presently grown varieties in that area.

Jefferson is a winter barley and was developed and released cooperatively with the Purdue University Agricultural Experiment Station. Jefferson is 6-rowed, awnleted, feed barley. Jefferson is resistant to leaf rust, powdery mildew and scald, and moderately resistant to net blotch. Like Harrison, its bearded sister selection, Jefferson, is winterhardy and has exceptional post-ripening standing ability. The principal advantage of Jefferson is its ease of threshing, when grown in the humid winter barley area, due to almost complete absence of awns.

The winter feed variety Miller was cooperatively developed and released with the Georgia Agricultural Experiment Stations. Miller was developed specifically to provide a leaf rust-, powdery mildew-resistant winter barley for Georgia. It is also resistant to lodging and it appears to be adapted to the Upper Coastal Plain and Piedmont areas of Georgia. Miller is equal or superior in yield to other adapted varieties in this area, and will yield considerably higher under severe leaf rust epiphytotics.

4. Straw strength. Progress is being made at Beltsville, Md., and in Arizona, Idaho, and Montana by crossing adapted varieties with others having shorter and stiffer straw. Greater straw strength is urgently needed to support the added yield due to hybrid barley. The varieties Jotun, Belownee, T5-7c, and Jaydee are being used extensively for this purpose.

B. Physiology and Culture

- 1. Winterhardiness. Histological comparisons of ice structure in peripheral crown tissues were consistent with relative hardiness of winter barley varieties tested. In experiments at East Lansing, Mich., the ice structure was consistent with the activity of certain water soluble cell wall polymers. These substances act as inhibitors of freezing kinetics, and interfere with the rate of ice crystal growth. Stability of interaction with an ice lattice was found to determine the activity of the inhibitors studied.
- 2. Aluminum tolerance. A method was developed at Beltsville for classifying individual plants in nutrient solution for reaction to aluminum toxicity.

RPA 208 - CONTROL OF DISEASES

- 1. Barley yellow dwarf virus particles observed in host phloem. At Brookings, S. Dak., electron microscopy revealed that some cells in the phloem area of a vascular bundle of plants infected with barley yellow dwarf virus contain viruslike particles. These particles have been observed and recorded in tissue samples from young leaves, old leaves, and roots. Generally, they are found in only one or two cells but in these cells they are frequently so numerous as to form a solid mass filling the lumin of the cells.
- 2. Ribonuclease in barley stripe mosaic virus-infected plants. A decrease in chlorophyll was associated with an increase in virus concentration and ribonuclease in North Dakota. The virus concentration was highest about two days prior to the peak of ribonuclease activity. Leaves showing the most severe virus symptoms contained the most ribonuclease which was about twice as abundant in susceptible plants as in resistant plants.
- 3. Fungicides may affect barley malt quality. At Madison, Wis., when barley plants were sprayed with maneb, nitrogen was reduced, kernel weight and size increased, and seed color improved to increase malting quality. However, when plants grown at Carrington. N. Dak. were sprayed with zineb and with Dithane S-35, there were no effects on these factors and no effect on malting quality.
- 4. Systemic fungicide continues to control loose smut of barley. The oxathiin compound (2,3,dihydro-5-carboxanilido-6-methyl,4 oxathiin) again gave complete control of loose smut of barley at Beltsville. The fungicide is compatible with organic mercury fungicides.

5. Genetics of pathogenicity of Helminthosporium sorokinianum. The progeny from a cross between a virulent and an avirulent culture differed significantly in virulence at Raleigh, N. C. It was concluded that the virulence was controlled by genes at more than a single locus.

RPA 207 - CONTROL OF INSECT PESTS

- 1. Cereal leaf beetle. F_2 progenies of barley crosses involving two resistant lines (C.I. 6671 and C.I. 6469) were exposed to cereal leaf beetle in the field in Michigan. More resistant lines were found in progenies involving C.I. 6469 than those with C.I. 6671 as a parent.
- 2. Greenbug. The major effects of greenbug feeding appear to be localized in infested barley leaves. Cumulative amounts of greenbug feeding produces a quantitative, detrimental decline in chlorophyll content and the rate of photosynthesis, and an increase in respiration. At Brookings, S. Dak., leaves of resistant plants, in responding to feeding damage, maintained a higher chlorophyll content and a higher rate of photosynthesis. Electron microscope examinations have revealed that in damaged cells dark granular material accumulates in the vacuole of the phloem elements and also between the cells. Here it causes a thickening of the cell wall. Chloroplasts contain greatly increased amounts of lipids but other cell organelles are not affected.

RPA 405 - PRODUCTION OF FIELD CROPS WITH IMPROVED CONSUMER ACCEPTABILITY

A. Malting Quality

- 1. Influence of smut infection on barley and malt quality. Larker barley with high (22 percent) smut infection showed a slightly higher tendency for water sensitivity than a lower infected (2 percent) sample of the same variety. An oxathiin fungicide had little effect on either sample. There appears to be no deleterious effect of high smut infection on malting and malt quality.
- 2. Malting procedures reviewed. There was excessive growth of rootlets in the two germinators with rotating containers. By increasing the rotation rate of these containers from 40 rph to 105 rph rootlet development was reduced. It was found that when openings per chamber were standardized at 24 for the 225 gram (dry basis) samples, aeration was improved. Preliminary studies showed that germination time could be reduced to 5 days without loss or precision of evaluation.
- 3. Biochemical factors that determine quality. Two peptide hydrolases (A and B) from malt were partially purified and characterized for stability, extremes of pH and temperature, metal ion activation, kinetic constants, substrate specificity and proteinase activity. Both of these enzymes increase in the embryo during germination -- hydrolase A during the first 30 hours and hydrolase B over the entire perioa. Work has been started on the development of methods for evaluation of hybrid barley enzymes.

Three malt proteinases which hydrolyze hemoglobin were partially purified. Activity during extraction of the proteinases from green malt and subsequent treatments can be maintained at pH 4 to 5 with a rigorous exclusion of oxygen by reducing agents, and a nitrogen atmosphere for all solutions.

4. Quality evaluation. Approximately 3,300 samples from the 1966 barley crop were received and analyzed; about 2,300 were malted. In general, as a result of the poor filling of many samples, higher than normal nitrogen contents were found. Promising winter barley selections from Michigan and New York appeared better than or equal to the variety Hudson. Spring barleys grown at Ithaca, N. Y. showed good quality. Two-rowed spring barleys from Michigan were better than or equal to Betzes. Good quality was present in six-rowed selections from Wisconsin, including two short-strawed selections. Two-rowed selections from Washington were good enough to justify release as varieties. Outstanding quality was evident in several selections from Idaho.

About 40 samples were evaluated by pilot malting and brewing, and 80 samples were tested by micro-brewing. Washington 7698-62 was most promising and Wisconsin X1068-1 had acceptable brewing properties.

Publications - USDA and Cooperative Program

-- Barley --

RPA 307 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY

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RPA 208 - CONTROL OF DISEASES

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CORN AND SORGHUM BREEDING, GENETICS, DISEASES, PHYSIOLOGY, AND CULTURE

Crops Research Division, 183

USDA and Cooperative Program

	:		S	cie	ntist	Ма	n-Yea	rs	F.Y.	196	8		
Location of	•			Res	earch	Pr	oblem	Az				:	Total
Intramural Work	: 307	:	208	:	207	:	405	:	304	:	204	_:	
	•	:		:		:		:		:		:	
Corn	•	:		:		:		:		:		:	
Maryland	•	:		:		:		:		:		:	
(Beltsville)	: 1.9	:	-	:	-	:	-	:	-	:	-	:	1.9
Florida	: 1.0	:	-	:	-	:	-	:	-	:	-	:	1.0
Georgia (Tifton)	-	:	-	:	1.0	:	-	:	-	:	-	:	1.0
Illinois	-	:	-	:	-	:	2.0	:	-	:	-	:	2.0
Indiana	-	:	0.2	:	-	:	0.3	:	_	:	-	:	0.5
Iowa	: 1.2	:	0.3	:	0.5	:	-	:	-	:	-	:	2.0
Mississippi	: 0.9	:	1.8	:	0.3	:	-	:	-	:	-	:	3.0
Missouri	: 4.8	:	0.6	:	0.6	:	-	:	-		-	:	6.0
North Carolina	: 1.4	:	0.6	:	-	:	-	:	-	:	-	:	2.0
Ohio (Wooster)	: 1.4	:	1.4	:	-	:	-	:	-	:	-	:	2.8
South Dakota	-	:	-	:	0.6	:	-	:	-	:	-	:	0.6
Tennessee	: 0.7	:	0.2	:	0.1	:	-	:	-	:	-	:	1.0
Wisconsin	: -	:		:		:		:		:		:	
Total Corn	: 13.3	:	5.1	:	3.1	:	2.3	:	_	:	-	:	23.8
		:		:		:		:		:		•	
Sweet Corn	•	:		:		:		:	. 1	:		:	- 0
South Carolina		<u>:</u>		:		:		:	0.4	:	0.4	:	0.8
Total Sweet Corn	-	:	-	:	-	:	-	:	0.4	:	0.4	:	0.8
		:		:		:		:		:		:	
Sorghum		:		:		•		:		:		•	
Kansas (Hays)	: 1.0	:	-	:	-	:	-	:	-	:	-	•	1.0
Kansas (Manhattan):		:	1.3	:	-	:	-	:	-	:	-	:	2.0
Nebraska	: 0.9	:	0.1	:	-	:	-	:	-	:	-	•	1.0
South Dakota	-	:	-	:	0.6	•	-	:	-	:	-	:	0.6
Texas :	1.0	:	_	:		:		:		:	-	:	1.0
Total Sorghum		:	1.4	:	0.6	:	_	:		:	_	:	5.6
Total	: 16.9	:	6.5	:	3.7	:	2.3	:	0.4	:	0.4	:	30.2

Intramural program is supplemented by extramural support representing (a) 3.6 SMY's at State Agricultural Experiment Stations 1/, and (b) P.L. 480 funds in 2 countries representing 218,700 U.S. dollars equivalent. 1/ RPA 307, 1.8; RPA 208, 0.2; RPA 207, 0.9; and RPA 405, 0.7.

Major Cereals Project in Africa 2/

	:	Scientist Man-Years F.Y.	1968	
Location of Work	:	Research Problem Area	:	Total
	: 60	3:	:	
	:	•	:	
Corn, Sorghum, and Millet		•	:	
Kenya (Kitale)	: 1.	0:	:	1.0
Kenya (Nairobi)	: -	:	:	-
Nigeria (Ibadan)	: 1.	0:	:	1.0
Nigeria (Samaru)	: 4.	0:	:	4.0
Uganda (Serere)	: 3.	0:	:	3.0
Total Corn, Sorghum,	:	•	:	
and Millet	: 9.	0:	•	9.0
Total	: 9.	0:	:	9.0

^{2/} Research in Africa under the Major Cereals Project is in cooperation with the Agency for International Development, East African Agricultural and Forestry Research Organization, and Organization of African Unity, Scientific, Technical and Research Commission.

Problems and Objectives

Corn and sorghum remain our primary feed grains, as well as providing the source of important industrial products. These two crops have an annual value of approximately 4.5 billion dollars. Increasing yield losses are resulting from the corn rootworm, the southwestern stalk borer, the several corn viruses and other insect and disease pests. The development of resistant types or other effective control measures are urgently needed. Basic research in genetics, physiology, and pathology is required to maintain current levels of productivity and quality and to develop types having improved industrial and nutritional potential.

Major objectives of the research are to:

- 1. Develop more efficient corn breeding procedures.
- 2. Identify sources of resistance to the major disease and insect pests.
- 3. Study yield variation in relation to physiological processes.
- 4. Explore biosynthesis of oil, protein, and carbohydrates.

Progress - USDA and Cooperative Program

-- Corn --

RPA 307 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY

A. Breeding and Genetics

- 1. New inbred lines released. Six inbred lines were released. Mol8W (Missouri) and Oh514 (Ohio) are highly resistant to maize dwarf mosaic virus. B65cms (Iowa) is the cytoplasmic male sterile version of B65 which possesses resistance to both the first and second generation broods of the European corn borer. B68 (Iowa) is resistant to the European corn borer and possesses some resistance to northern leaf blight. B67 (Iowa) is moderately resistant to the western corn rootworm. B66 (Iowa) has good combining ability and is somewhat resistant to both root and stalk lodging.
- 2. Gene action in heterosis. The gene action involved in heterosis continues to receive attention. Quantitative genetic studies conducted at Raleigh, N.C., demonstrated significant epistatic effects in single, three-way, and double cross comparisons as well as for F_1 and F_2 contrasts. Grain yield, ear number, and ear height exhibited epistasis in sets of both random and selected lines. Although generally significant, epistasis accounted for only about 10 percent of the total variation for any given trait indicating additive and dominance effects to be of major importance.
- 3. <u>Mutant poller</u>. Techniques were developed at Missouri for handling aqueous suspensions of corn pollen. This provides a simple procedure for the application of mutant pollens or for differential selection tests

utilizing known heterozygotes. Preliminary results have been encouraging.

Several flavonoid type compounds have been identified from pollen, quercetin and isoquercitrin. The finding of these two compounds led to the discovery that pollen has an enzyme system for converting quercetin to its 3-glycoside, isoquercitrin. Further studies using mutant pollens led to the establishment of the gene-enzyme relationship, Bz₁ locus-glycosylase enzyme.

- 4. Overlapping translocations. Attempts to obtain desirable genes in duplicate have been conducted at Missouri through the use of overlapping translocations. Results thus far have been largely negative through failure to maintain the duplicated chromosome fragment.
- 5. <u>Isozymes</u>. Thin layer chromotography studies at Raleigh, N.C., have indicated the presence of several peroxidase isozymes. Several loci, each with two or more alleles, appear to be involved. Isozymes produced by certain loci are tissue specific; being present in certain tissues and absent in others of a single plant. Some of these isozymes appear to play a role in the regulation of indoleacetic acid activity.

B. Physiology and Culture

1. <u>Mineral nutrition</u>. Studies in Ohio concentrated on the changes in organic acids associated with the expression of mineral deficiency symptoms. The predominant organic acid of normal corn leaves is t-aconetic. In calcium, potassium, magnesium, and phosphorus deficient leaves t-aconetic acid decreased and was accompanied by a marked increase in malic and citric acids. Studies on the synthesis and degradation of these acids may shed light on the function of the major nutrients in the physiologic processes.

RPA 208 - CONTROL OF DISEASES

- 1. Stunt virus. One of the four known viruses of corn causes stunt. Studies in Mississippi indicate that stunt continues to spread in the Southeastern States. In this region the known insect vector, <u>Dalbulus maidis</u>, is not considered to be the effective one. Other leaf hoppers are suspected vectors, but evidence for their transmissive ability is not yet conclusive. Corn acreage has decreased markedly in areas of heavy stunt infection, and relatively few lines possess adequate resistance.
- 2. <u>Maize dwarf mosaic</u>. Two strains of maize dwarf mosaic have been identified in Ohio and the Northeastern States. The more serious and widely distributed strain, A, occurs in areas where Johnsongrass is common and this plant serves as the overwintering host. The second strain, B, occurs where Johnsongrass is not found and will not infect Johnsongrass. Good sources of resistance are available for both strains, A and B.

- 3. Wheat streak and other viruses. In corn, wheat streak mosaic causes losses in Ohio, northern Indiana, Michigan, and adjacent areas. It is not common in Nebraska, however, where wheat streak mosaic is a common virus on wheat. This suggests that the strain which infects corn may be different from the wheat strain or that there may be a strain difference in the vector, the wheat curl mite. Feeding injury from this mite is also responsible for "kernel red streak." An additional virus which is mechanically transmissible and for which Johnsongrass serves as an overwintering host is widespread in the southeast. Symptomology is quite different from that for maize dwarf mosaic and the resistance -- susceptibility pattern is dissimilar. This may represent a third strain of maize dwarf mosaic.
- 4. Fungus diseases. Techniques have been developed in Mississippi for the evaluation of resistance to Fusarium as a seedling blight. Screening for resistance is in progress.

Fusarium moniliforme and Gonatobotrys zeae were frequently associated with seed from virus-infected corn plants. G. zeae is a new species which has been described on corn. Apparently the invasion of corn by viruses predisposes it to heavy attack by F. moniliforme. This is the most common fungus found on corn seed and it causes poor stands. It also causes ear rots. Other fungi causing poor stands are Pythium sp. Helminthosporium spp., Penicillium sp. and Aspergillus sp.

Downy mildew caused by <u>Sclerospora sorghi</u> continues to spread in Mississippi, especially on forage sorghums where an abundance of spores are produced. In fields where corn follows sorghum in rotation, downy mildew has presented a problem. The season of the year appears to have an effect on the symptom expression of downy mildew.

RPA 207 - CONTROL OF INSECTS

1. European and southwestern corn borers. In studies conducted in Iowa, resistance to first generation leaf feeding by the European corn borer has been associated with a cyclic hydroxamate, the compound DIMBOA (2, 4-dihydroxy-7-methoxy-1,4-benzoxazine-3-one). The biosynthetic pathway of this and related materials and the nature of inheritance are being investigated under a grant. Recurrent selection has been established as an effective procedure for increasing the level of resistance in breeding populations. Limited tests conducted in Mississippi, involving artificial infestation with southwestern corn borer egg masses, have failed to reveal any important differences in resistance to this important pest. These tests indicate, however, that the genes for resistance to southwestern corn borer are different from those conditioning resistance to the first and second brood of European corn borer.

- 2. Corn earworm. The nature of resistance to the corn earworm is being studied under a cooperative agreement in Missouri. Improved techniques are being developed for detecting growth stimulators, suppressors, or toxins in silks which affect earworm larvae. A diet-dip bioassay technique was devised whereby limited amounts of silks can be effectively tested. Related studies on inheritance are conducted at the Southern Grain Insects Research Laboratory at Tifton, Ga. Recurrent selection has been effective in increasing the average level of resistance. In the tests at Tifton, a corn earworm resistant line, 245, was found to carry genes for resistance on chromosomes I, IV, V, IX, and X. Diallel tests have established that gene action is largely additive but that dominance and epistasis are also involved.
- 3. Western corn rootworm. Studies on resistance to the western corn rootworm are being conducted at the Northern Grain Insects Research Laboratory at Brookings, S. Dak., and at Columbia, Mo. A few lines have been identified that possess some degree of resistance to or tolerance of rootworm feeding.

RPA 405 - PRODUCTION OF FIELD CROPS WITH IMPROVED CONSUMER ACCEPTABILITY

1. Protein synthesis. The three nucleases in corn have been further purified and characterized under a cooperative agreement with Illinois. RNase I is soluble and found in large amounts in the developing endosperm. It possesses RNA and DNA activity and possibly also 3'-nucleotidase activity. RNase activity is inversely correlated with protein percentage; types such as Illinois High Protein have low RNase activity and low protein types have high activity. The opaque-2 gene, when homozygous, gives rise to a marked increase in RNase. RNase II is similar to RNase I except it occurs in a microsomal fraction.

Reciprocal crosses involving Illinois High Protein and three inbred lines revealed that the RNase and protein content and the amino acid composition were similar to the maternal parent. Protein body preparations from developing endosperm were low in zein, thus differing from the protein of the mature seed.

2. Oil synthesis. Studies conducted at Illinois indicated that lipid synthesis is most active during the period from 15 to 45 days after pollination. In Illinois High Oil, however, synthesis may continue for periods up to 90 days. Each lipid class has a characteristic fatty acid composition. The rate of acetate - 14°C incorporation into fatty acids increased with age during the kernel development period. In crosses involving Illinois High Oil the maternal parent had a pronounced effect on germ size, oil content, and fatty acid composition.

Under a cooperative agreement with Illinois, techniques have been developed for the fatty acid analysis of single kernels and of embryo plugs without destroying the value of the kernels for planting. It has been strongly indicated that differences in palmitic acid among strains of corn may be due to more than a single gene. Strains also have been found that are different in myristic acid content, however, nothing yet is known regarding the inheritance of this fatty acid.

3. Carbohydrate synthesis. The fate of translocated sugar in normal kernels and in the endosperm mutants, ae, du, sh2, su1, su2 was studied in Indiana. The use of \$14CO_2\$ indicated radioactivity was first apparent in the free glucose and fructose sugars. Later \$14C\$ in sucrose increased and that in the monosaccharides decreased. Prior to or during translocation to the developing kernel the sucrose is hydrolized to simple sugars. After entry into the endosperm the simple sugars are synthesized into storage carbohydrates, the type and amount being determined by the genetic constitution of the plant. Procedures have been developed for the quantitative separation of each of the main carbohydrates. The enzyme systems necessary for the synthesis of storage carbohydrates may be bound to the membrance of the amyloplasts.

-- Sweet Corn --

RPA 304 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY

1. Mutant genes improve sweet corn. F_1 hybrid sweet corn with endosperm mutant genes <u>ae</u>, <u>du</u>, and <u>wx</u> were produced at Charleston, S.C. Carbohydrate analyses and taste tests show the <u>ae du wx</u> genotype will provide more palatable corn for human use than the standard <u>su</u> type. The mutant hybrid contained 7.5 percent total sugar (fresh weight) compared to 4.5 percent for standard sweet corn; quality also held better during postharvest handling.

Two hybrids and two inbreds show good tolerance to both the stunt virus and maize dwarf virus.

RPA 204 - CONTROL OF INSECTS

1. <u>Insect resistance</u>. The factor or factors that result in corn earworm larval mortality up to 30 percent, and retarded growth of survivors on resistant sweet corn, apparently do not significantly affect other stages of the insect. Days in pupal stage, pupal and adult mortality, and eggs per female did not appear to be related to resistance in South Carolina tests.

A technique of extraction, chromatographic separation, and bioassay with earworm larvae feeding on artificial media appeared to be a reliable method for detecting resistance factors in sweet corn silks.

-- Sorghum --

RPA 307 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY

A. Breeding and Genetics

- 1. New inbred lines released. Two inbred lines were released from Nebraska. R-NB6250 is a restorer line used as the male parent in the grain sorghum hybrid RS633. The related line R-NB6229 is used as the male parent in the production of the forage hybrid NB306F.
- 2. Chromosome linkage. The 10 primary trisomes of sorghum have been identified in Texas. Over 100 homozygous translocation stocks have been verified. Intercrosses among these and with the trisomic stocks have led to an arbitrary designation of chromosomes. Considerable progress has been made in relating these chromosomes to known linkage groups.
- 3. World collection of sorghum and of Pennisetum. Under PL 480 projects in India, detailed descriptions of this material have been published, and the collections have been developed and properly stored in New Delhi, India. They are available to Federal, State, and private breeding programs in the United States.
- 4. Quantitative genetics. The genetic studies have been conducted at both Manhattan, Kans., and Lincoln, Nebr., in extramural research activities. The work at Lincoln involves the development of random mating populations through the use of genic sterility. These populations will be utilized for the evaluation of relative efficiencies of several types of recurrent selection schemes. The work at Manhattan has utilized selected material to estimate heterosis for yield and its components and for studies on inbreeding depression. In measuring genetic variability or stability of pure lines, data have shown that epistatic gene effects cannot be neglected, especially the aa and dd types.

B. Physiology and Culture

- 1. Heat and drought stress. At Lincoln, Nebr., drought and heat tolerances were not found to be highly correlated. Lethal temperatures for millets are generally higher than those for sorghum. One drought-resistant corn selection (Conoco) also had a higher heat tolerance than some of the sorghums. Sorghum, however, generally maintains a lower water potential than either millet or corn.
- 2. Photosynthesis in field grown plants. In Nebraska, photosynthesis of entire plants regularly exhibited two maxima; one was at 12:30 p.m., after which there was a decrease and then another was at 4:30 p.m. The midday decrease is presumed to arise from some type of stress.

3. Nitrogen nutrition and protein synthesis. Activities of the two enzymes, glutamic dehydrogenase and glutamate-oxalacetate-transamenase, involved in protein synthesis were determined in compact and semiopen inflorescences in Nebraska. These enzymes are most active at anthesis and their activity gradually declines until the milk stage. This is followed by a second increase in activity and then a gradual decline until the kernels are physiologically mature. Semiopen heads had higher activities than compact heads through all the tests and thus they may be considered to be more effective for photosynthesis.

Labeled ¹⁴CO₂ was used to measure the photosynthate translocated to the maturing grain; Leaf 3 and the flag leaf were selected in which to introduce the ¹⁴CO₂. The flag leaf was the greater contributor even though it was about one-third the size of the third leaf. This points out the importance of the flag leaf. Preliminary studies concerning translocation pathways indicate that the isotope moved from midleaf to the stalk in less than 20 minutes.

RPA 208 - CONTROL OF DISEASES

- 1. Smut. Most hybrids in commercial production possess a satisfactory level of resistance to the five known races of kernel smut. Head smut continues to spread throughout the Great Plains area. Sources of resistance have been found and resistance is rapidly being incorporated into parents of commercially acceptable sorghum hybrids.
- 2. <u>Downy mildew and virus</u>. Two new diseases are becoming increasingly important: downy mildew and maize dwarf mosaic. Many of the widely used parents are susceptible to maize dwarf mosaic. Sudangrass and many of the forage sorghums are highly susceptible to downy mildew. Second cutting yields are seriously reduced for the sudan-sorghum hybrids observed.
- 3. Charcoal rot. This disease continues to cause serious yield reductions and stalk lodging under certain environmental conditions. Techniques for artificial inoculation have been developed and evaluated under a contract with Arizona. Growing plants under water stress is a valuable technique in the screening process. Strain differences in susceptibility exist but no types have been identified which possess a high level of resistance.

-- Major Cereals Project --

RPA 603 - TECHNICAL ASSISTANCE TO DEVELOPING COUNTRIES

Research activities of the major cereals project are fully cooperative with and form a part of the total research erfort at a given location. Research centers are established in both East Africa and West Africa. One activity center is in Nigeria, one in Uganda, and one in Kenya. Several locations make up each center.

- 1. Regional sorghum trials. Research activities of the entire program are fully cooperative and form a part of the total effort. Cooperating countries in the initiation of this first regional sorghum nursery in West Africa are Cameroon Republic, Chad, Dahomey, Ghana, Niger, Nigeria, Senegal, and Upper Volta. The 24 items included represented entries from India, United States, and Uganda as well as from the cooperating countries. The first sorghums of the Nigeria hybridization program were included.
- 2. Sorghum trials in Northern Nigeria. Small scale experimental seed production trials were initiated. In these plots, the potential value of earlier maturing, short-stalked varieties was demonstrated for this area of limited rainfall. The U.S. variety Hegari gave good yields on the irrigated scheme, however, this variety is not preferred as food by the Nigerians.
- 3. <u>Male sterile lines developed</u>. Cytoplasmic sterile and fertility restorer lines of sorghum with the proper maturity have been developed at the Institute for Agricultural Research in Northern Nigeria. Trials indicate a three-fold increase in yield potential.
- 4. <u>Sorghum hybrids</u>. At Serere, Uganda, sorghum hybrids have produced over 7,000 kilos per hectare. Resistance to <u>Helminthosporium turcicum</u> and <u>Colletotrichum graminicolum</u> has been improved. Tetraploid stocks continue to show promise.
- 5. Fertilizer trials. In Uganda, fertilizer trials with sorghum, bullrush millet, and finger millet indicate limited response to nitrogen or phosphate when either is applied singly. Yield can increase 50 percent or more when a combination of the two fertilizers are used.
- 6. <u>Maize breeding in Northern Nigeria</u>. A program was initiated in which the parental materials involved both local varieties and introductions from Colombia, South America.
- 7. Regional maize trials. West Africa Regional maize trials were grown in eight countries: Cameroon Republic, Dahomey, Ivory Coast, Liberia, Nigeria, Senegal, Sierre Leone, and Upper Volta. Work is continuing on the development of broad-based populations. The high lysine gene, opaque-2, is being introduced into these populations. New sources of resistance have been found for leaf rust (Puccinia polysora) and leaf blight (Cochliobolus heterostrophus).
- 8. <u>Maize seed production</u>. In Kenya, maize seed production appears to be safely above local needs. In addition to meeting the demands of Kenya, seed for approximately 7,000 acres of Kenya developed hybrids have been sold for growing in Tasmania.

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-- Corn --

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RPA 208 - CONTROL OF DISEASES

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-- Major Cereals Project --

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WHEAT AND RYE BREEDING, GENETICS, DISEASES, OUALITY, PHYSIOLOGY, AND CULTURE

Crops Research Division, ARS

USDA and Cooperative Program

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Location of Intramural Work		: Research Problem Area :										
	:	307	:	208	:	207	:	405	:			
	:		:		:		:		:			
Wheat	:		:		:		:		:			
Maryland (Beltsville)	:	2.5	:	1.7	:	-	:	0.3	:	4.5		
Idaho	:	1.0	:	-	:	-	:	_ `	:	1.0		
Kansas	:	-	:	2.3	:	-	:	3.7	:	6.0		
Michigan	:	-	:	-	:	0.4		-	:	0.4		
Minnesota	:	0.5	:	4.2	:	-	:	_	:	4.7		
Missouri	:	0.8	:	0.2	:	-		-		1.0		
Montana	:	0.4	:	0.3	:	0.2	:	0.1		1.0		
Nebraska	:	0.5	:	1.9	:	0.3	:	0.3	:	3.0		
North Dakota	:	1.4	:	1.0		0.5	:	2.0	:	4.9		
Ohio	:	-	:	-	:	-	:	2.0	•	2.0		
Oklahoma	•	-	:	1.0	:	-	:	_	:	1.0		
Oregon	:	0.2	:	1.8	:	-	•	-	:	2.0		
South Dakota	:	-	:	-	:	0.6	:	-	:	0.6		
Texas	:	0.8	:	1.7	:	_	:	_	•	2.5		
Washington	:	2.3	:	3.0		_	:	3.0	:	8.3		
Total Wheat	:	10.4	:	19.1	:	2.0		11.4		42.9		
	:		:		:		:		:			
Rye			:		:		:		:			
Maryland (Beltsville)	:	0.2	:	-	:	-	:	-	:	0.2		
Total Rye	:	0.2	:	-	:	-	:	-	:	0.2		
Total	:	10.6	:	19.1	:	2.0	:	11.4	:	43.1		

Intramural program is supplemented by extramural support representing (a) 2.9 SMY's at State Agricultural Experiment Stations 1/, and (b) P.L. 480 funds in 4 countries representing 83,043 U.S. dollars equivalent.

^{1/} RPA 307, 0.6; RPA 208, 1.0; RPA 207, 0.3; and RPA 405, 1.0.

Problems and Objectives

Wheat is extensively grown in the United States and is widely used as human food. Practices that enable farmers to produce a better crop at lower cost are the main objectives. Higher yields, more resistance to hazards, and more nutritious grain are sought. A number of questions need to be answered and urgent biological problems present themselves. Some of the problems involve the factors and the types of gene action in heterosis; the mechanisms of sterility and restoration of fertility; the mechanisms of mutation and/or gene interaction that go to produce new and high levels of nutrition; and the stability of improved quality. New sources are needed for resistance to diseases and insect pests. Each disease and pest should be investigated thoroughly to discover ways to reduce damage. Rye as a grain and as a forage crop is low in yield and is threatened by certain diseases. Triticale trials and breeding improved lines are needed. All of these problems have a far reaching impact on food production, quality, nitrogen, and economic utilization of wheat and rye.

Major objectives of the research are to:

- 1. Increase the national average yield to 30 bu. per acre (about 20 percent).
- 2. Eliminate epidemic disease losses.
- 3. Develop resistance barriers to insect outbreaks.
- 4. Improve physiological mechanisms and cultural management to mitigate environmental stresses.
- 5. Modify flour composition for better nutrition and processing quality.

Progress - USDA and Cooperative Program

-- Wheat --

RPA 307 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY

A. Breeding and Genetics

1. New varieties released -- grouped by market classes. SOFT RED WINTER-Arthur (C.I. 14425) was released in cooperation with the Purdue University Agricultural Experiment Station and the Entomology Research Division. It is high in productivity and milling quality and combines some of the best resistance to stem rust and loose smut of any wheat available for use in the Ohio Valley. It is moderately resistant to other common diseases although some leaf rust cultures attack it and it is not resistant to certain new races of hessian fly.

Timwin (C.I. 13787) was developed in cooperation with the Wisconsin Agricultural Experiment Station. It is a semi-dwarf variety moderately resistant to lodging. Timwin is resistant to bunt and moderately resistant to leaf and stem rust. It has soft wheat quality equal to Knox.

HARD RED WINTER - Crest (C.I. 13880) was developed and released in cooperation with Montana Agricultural Experiment Station. It embodies a high type stripe rust resistance from P.I. 178383 of the USDA World Collection. This parent also has excellent resistance to bunt. Crest is well adapted to western Montana.

Trader (C.I. 13998) and Trapper (C.I. 13999) were developed in cooperation with the Nebraska Agricultural Experiment Station. The Colorado Station joined in the release. These two varieties are similar in characteristics and surpass other varieties in stem rust resistance. Trapper appears to be more winterhardy and will fit a special area requiring more hardiness. It is expected that Trader will have rather wide adaptation in western Nebraska and adjoining portions of nearby States. Both have excellent milling and baking characteristics.

Shawnee (C.I. 14157) was developed cooperatively with the Kansas Agricultural Experiment Station. It resembles Ottawa in general characteristics but has exceptionally high quality among hard red winter wheats. Its adaptation is primarily to eastern Kansas and adjacent areas of adjoining States.

HARD WHITE - Adams (C.I. 13722) was released with the Oregon Agricultural Experiment Station. The variety has medium plant height, moderarely stiff straw and is productive on both dry land and irrigated fields, especially in eastern Oregon. It is resistant to most races of bunt and has tolerance to stripe rust. It has excellent milling properties and the flour is a strong-gluten type.

HARD RED SPRING - Polk (C.I. 13773) was developed cooperatively with the Minnesota Agricultural Experiment Station. Polk has excellent test weight and milling and baking quality. Its broad rust resistance base and wide adaptation make it one of the best varieties ever released from the cooperative project. It was jointly released by ARS, Minnesota, Montana, North Dakota, and South Dakota.

- 2. New germ plasm released. In cooperation with the Texas Agricultural Experiment Station wheat germ plasm having possible new fertility restoring genes was released to breeders for use in developing hybrid wheat. It is based on fertility restoration from a composite of diverse germ plasm.
- 3. Genes mapped with aid of telocentrics. Two genes were mapped with the aid of telocentrics at Columbia, Mo. The Thatcher stem-rust gene <u>Srl6</u> on the long arm of chromosome 2B is at least 50 crossover units from the centromere. The Axminster mildew-resistance gene <u>Pml</u> on the long arm of chromosome 7A is also 50 or more units from the centromere.
- 4. A line homozygous for resistance to both <u>Lr9</u>, leaf-rust, and <u>Srll</u>, stem-rust resistance. In this line obtained from Columbia, Mo., resistance to leaf rust is due to a chromosome segment transferred from Aegilops umbellulata; the stem rust resistance is from Timstein variety of wheat. The line is

homozygous for <u>Ki</u> (Pollen Killer), and will, therefore, provide for excellent transmission of the resistance genes following crosses. Existence of a neutral allele at the Ki locus was confirmed for the varieties Yalta and Oasicolum.

5. Mutagens. Work was undertaken several years ago at Stillwater, Okla., to transfer resistance to wheat leaf rust from a wheat x Agropyron hybrid to common wheat by X-ray irridation. Twenty-two possible translocation stocks have been identified genetically. One of these has been verified cytogenetically. The first objective was to identify lines homozygous for the "Agropyron type" of leaf rust resistance at the mature plant stage. Among the 1,594 entries observed, 827 were resistant. A second goal was to induce translocation of the genetic material controlling resistance to wheat streak mosaic virus from a wheat x Agropyron hybrid to common wheat by way of material irradiated with X-rays or thermal neutrons. Of 270 hybrid families tested, 121 promising lines were saved for cytogenetic examination and virus trials.

Recurrent treatments of Little Club wheat with the chemical mutagen ethylmethane sulfonate (EMS) have been continued in North Dakota studies. The primary objective is to induce mutations for stem rust resistance. In a test of 56 progenies with culture lll-SS2 of stem rust, one progeny showed 18 immune seedlings (or escapes) and 2 susceptible, whereas 55 other progenies were heavily infected and susceptible. Recurrent treatment of Lakota durum with EMS was made to effect changes in kernel size and for mutants of possible agronomic value. About 70 plants with short stature were selected for further study. Within a group of 57 selections derived from recurrently X-irradiated Langdon that appeared to be highly vigorous, none appeared to yield much better than untreated checks.

- 6. Semidwarf and day-length non-sensitivity durums. Under the disease-free conditions of 1967 in North Dakota, several durum semidwarfs appeared promising, but so far, performance of most semidwarf durums has been unstable. Several lines only 3 to 5 inches shorter than Wells or Leeds showed considerable promise in all respects in both 1966 and 1967. A trial of durum selections differing in day-length response indicated that non-sensitive types were equal to sensitive (long-day) types in yielding ability. It appears that durum breeding lines can be converted to non-sensitivity without loss in productivity. Non-sensitive types would be more universally adapted than long-day types.
- 7. Semidwarf common wheats continue to gain prominence. In Minnesota, 26 advanced experimentals were semidwarf and 2 are known to be daylight insensitive. All lines possess good stem and leaf rust resistance. Almost all of the semidwarfs appear to have satisfactory test weights, a characteristic which is lacking in previous semidwarfs. Semidwarfs yielded 10 to 29.5 percent better than the variety Chris. Comparable progress is being made at seven other stations.

- 8. Hazard tolerance broadened in new types of wheats. Progress in breeding new varieties that are more hazard tolerant than Gaines is being made in Washington and Oregon. In the summary of yields from five of the regular white wheat nursery trials, commercial varieties grown prior to development of semidwarf were the lowest vielders of the 112 entries and were in the same order within themselves as commonly found a decade ago. Two semidwarfs, Gaines and Nugaines, ranked 49th and 32nd respectively. The four varieties which ranked highest and the two which ranked 16th and 24th in vield were selected for intensive agronomic and quality tests to determine which ones may be suitable for release. All six showed a high level of resistance to stripe rust in both the seedling and mature stages of plant growth. All vielded higher in the foot rot nursery than their standard height counterparts of comparable grain quality. However, the Burt type semidwarfs showed percentage losses from inoculations with foot rot similar to those of Burt. Suwon 92/4*Omar, a short standard height selection, represents an unexpected level of foot rot tolerance found in many other selections from the same cross. One of the six selections is shorter than Gaines and represents an unusual level of improvement in fast emergence of a short semidwarf for early seeding.
- 9. Hybrid wheat continues enticing but seed set is mediocre. In Washington, most hybrids have failed to yield equal to or above Sel. 101 (C.I. 13438). Of the 109 hybrids tested, 37 yielded from 0 to 61 percent more grain than Sel. 101 and 46 yielded 20 to 61 percent less. Forty-five hybrids produced grain yields equal to or superior to the highest yielding parent and 17 of them exceeded the best parent by 20 to 77 percent. No particular association occurred between heading date or tillering and the high yielding ability of the hybrids. Low test wieght continued to be a problem among the higher yielding hybrids. A majority of the hybrids had culm lengths taller than Sel. 101, whereas, 40 were equal to or shorter than this selection. The results of three years of "A" seed line production suggests that a 50 percent return on an "A" line such as Sel. 101 is feasible. In contrast, the relatively poor returns (18 percent) with "A" line Sel. 1 makes economical production questionable.

In Nebraska, the average yield of hybrid wheat was below that of the parents. The yield level of some of the hybrids, and perhaps all, were influenced by the degree of restoration. The Gage and Scout hybrids were closest to normal in male fertility and the Omaha hybrid was the farthest from normal. The hybrids performed relatively better when compared to the parental lines at Mead where growing conditions were the poorest than when they were at North Platte where conditions were not as severe. Comparisons between the standard variety used as the female parent and its hybrid counterpart show that the hybrids are as food, and in some cases, superior in quality to the standard variety. This provides considerable assurance that the quality of hybrids can be expected to be similar to the quality of varieties currently grown in this area. It should be pointed out that the restorer parent which is used has good inherent baking properties and high protein content. It should also be noted that the hybrids did not differ drastically in yield from their parents, although sometimes the difference was significant.

10. Isogenic pairs for plant height assess value of semidwarf wheats. Three years of data from Washington are available on 180 lines of the isogenic population, Sel. 14/7*Bart. They are subdivided into 3 groups of 60 lines each according to culm length, short, 14 to 15 inches; medium, 21 to 25 inches and tall, 29 to 34 inches.

Correlations between yield and culm length (height) were positive and highly significant for all crop years when the 3 height groups were pooled. When the 3 subgroups were considered separately, height and yield in the short and medium semidwarf groups were positively correlated but the r values were significant only for 1965. Tall lines tended to correlate highly with yield in all crop years, except 1967. In contrast, the yield/height relation of short semidwarfs was correlated most closely in 1967. These results demonstrate close, consistent, positive relationships between height and yield in the Sel. 14/7*Bart material when grown under annual cropping. This pattern holds true even when minor height differences on the order of 1 to 4 inches are involved. Hence, the value of semidwarfs is not implicit in their shortness.

Semidwarf types, susceptible to stripe rust, suffer greater losses in their yield than susceptible standard height lines. The yield advantage of semidwarf wheats can readily be lost when stripe rust becomes a limiting factor.

B. Physiology and Culture

1. Vernalization studied at Corvallis, Oreg. The vernalization process in Elgin winter wheat was not enhanced by cyclic temperature treatment (3 hr. at 5°, 1 hr. at 18°, 3 hr. at 5°, etc.). Approximately 700-800 hours of continuous or intermittent 5° temperature were required to satisfy the vernalization requirement of this variety.

Abscisic acid sprays in concentrations greater than 1 ppm had an inhibitory effect on meristem elongation of spring and vernalized winter wheat. The inhibiting effect on the vernalized winter wheat was much greater than on the spring wheat. Abscisic acid and chlorocholinechloride (CCC) interfere with the gibberellin metabolism and both chemicals reduce culm elongation. They do not interfere with the flowering response of wheat. These data indicate that substances other than gibberellins probably play a minor role in the floral initiation and development of wheat.

2. Storage of grain results in damage to some components. Damage to the breadmaking potentialities of stored wheat flour was accompanied by an almost complete breakdown of free flour lipids and by a substantial decrease of bound lipids in studies at Manhattan, Kans. Starch-gel electrophoresis patterns indicated that proteins of storage-damaged flour had undergone only minor changes. Mixing time of damaged flour was more than twice as long as that of sound flour. Gassing power of the damaged flour was comparable to that of the sound flour. When loaves were made from the damaged flour, volume potential and crumb color could be restored by adding polar or total lipids but color was not improved. Fractionation studies showed that the damage was due to the breakdown of the lipids rather than to changes in the gluten proteins or starch and water solubles.

- 3. Lipids change during kernel development. Free lipids decreased little during maturation of wheat at Manhattan, Kans. Bound lipids decreased markedly. Calculated on a kernel basis, there was a 3 to 5-fold increase in free lipids as the wheat matured. The changes were accompanied by a consistent and substantial increase in free polar lipids. No changes were found in the distribution of bound polar lipids. There was, however, a consistent decrease in the lipids in free fatty acids, and in mono- and diglycerides during maturation. A number of changes which included an increase in monogalactosyl glyceride in free polar lipids of maturing wheat were found.
- 4. Fertility trials. Heading date, height, test weight, and yield of 3 tall varieties and two semidwarf lines as influenced by soil application of 5 levels of nitrogen and 2 levels of phosphorus were evaluated in Minnesota. Added fertility had little or no influence on heading date and height. There was an inverse effect on test weight as it decreased about 1.5 lbs/bu. from the control level to the highest fertility level. Satisfactory bushel weights were obtained even at the high level. The largest response was observed in yield of the semidwarfs. The semidwarfs responded more favorably to nitrogen than the taller varieties. At the 51 lb. phosphorus and nitrogen level 1 experimental semidwarf yielded 35 percent more than Chris, a taller variety. This is consistent with last year's data.
- 5. Freezing trials separate hardiness levels among spring wheats. At Pullman, Wash., when field-hardened spring wheat crowns were exposed in a cold chamber, some of the wheats revealed the ability to harden. Others seemed to lack this property completely. A good correlation exists with field rankings to show significant varietal differences in winterhardiness.

RPA 208 - CONTROL OF DISEASES

A. Control of Rust and Mildew

- 1. Near-isogenic lines show value in mildew control. There was a 15 to 7 bu./A (25 percent) gain in yield of mildew resistant vs mildew-susceptible isogenic pairs of lines tested in 1968 at Tifton, Ga. When leaf rust was also controlled, there was an added gain of about 3 bu./A. These and near-isogenic pairs for several stem rust alleles are deposited in the World Collection.
- 2. Systemic oxathiin fungicides gave short-time rust protection. When DCMOD (Plantvax* F 461) was applied on Little Club wheat as a spray for leaf rust control there was an increase of: 39 percent spikes; 64 percent more seeds; and 16.4 percent heavier seeds than in non-treated plants. Spraying barley 1 to 2 times with DCMOD for leaf rust control resulted in yield increases of 10.6 percent to 177.8 percent.

- 3. Rust prediction data and models analyzed. Cumulative counts of rust spores caught on exposed surfaces of adhesive-coated rods tell a meaningful story of epidemic development. In Kansas, the method has been used in conjunction with weather records and crop development to predict the course and severity of epidemics. A linear multiple regression model for predicting numbers of spores of leaf and stem rust at different dates beyond a date of forecast was tested. Regression equations solved from spore count and weather data from the 1953 and 1961 to 1965 seasons were used. The log cumulative spore counts on the dates to be forecast were used as a dependent variables with good results of prediction for the spring wheat region.
- 4. Viability of rust cultures on artificial media. Australian Puccinia graminis tritici cultures maintained in a Minnesota laboratory for many months produced normal uredial infections in wheat leaves. Isolates representing 11 North American races of P. graminis tritici have not grown on the peptone medium, although the search for culturable North American isolates continues. Growth of the Australian rust fungus on a non-living medium represents a major breakthrough in the study of rust fungi.
- 5. Genetic studies on rust resistance. Isolation of lines monogenic for resistance to culture lll-SS2 from crosses of Spelmar, St464, Kubanka, and Iumillo with Marrocos 9623 was accomplished from F4 and backcross F3 families in North Dakota. Each chosen homozygous family had sib families which appeared to segregate for a single gene for resistance. F2 samples from Marrocos 9623 crossed with lines derived from C.I. 8155 x Marrocos 9623 indicated that each of ten lines was homozygous and monogenic for resistance to culture lll-SS2. Rust tests on seedlings of the F2 and the F1 backcross of Beladi 116 x Marrocos 9623 indicated that Beladi 116 has three dominant genes for resistance to culture lll-SS2 of stem rust. The number of genes in lines derived from Kota x Little Club which condition resistance to culture lll-SS2 and their allelic relations to genes for resistance in lines derived from Marquis and Reliance indicate that one line derived from Kota, and designated Kt-A, had at least two genes for resistance.

The hexaploids, Justin and Little Club, were crossed with the tetraploids Leeds, Ramsey, Stewart 63, and Sel. 56-1, to transfer leaf and stem rust resistance from the tetraploids to the hexaploids. Results indicate that Sel. 56-1, Stewart 63, and Leeds differ from Justin and Little Club by at least two genes for leaf rust resistance. Ramsey may differ from Justin and Little Club by one gene. Resistance appeared to be recessive in all crosses.

6. Stripe rust resistance in P.I. 17883 linked with brown glume color. Plants that are heterozygous for rust resistance and glume color occasionally develop rust susceptible sectors (leaves and glumes that are variegated or white). In reciprocal backcross and test cross populations in Oregon, mutants were found in populations representing F1's of (P.I. 178383 x Orin). The F1 can be used as either the female or male in the test cross. One mutant was recovered for every 184 gametes tested. Indications were that a major gene for stripe rust in P.I. 178383 is linked with brown glume color.

7. Rust physiology. At equivalent levels of inoculum, leaves which showed rust resistant reactions in Nebraska gave rise to esentially the same percentage increases in decarboxylation rates as those for the susceptible infected plants during the first 2 or 3 days after infection. Subsequently, the rates of decarboxylation for resistant infected tissue increased to values that were approximately 8 times as great as those for healthy tissue of comparable age, even though visible signs of infection were negligible. The rates were first measured at the stage of disease development which correlated with the onset of resistance and were observed for at least 10 additional days.

Studies of stem rust spore metabolism was limited because consistently high levels of germination en masse with subsequent formation of appressoria, infection pegs, and substomatal vesicles was hard to get. Results of studies to devise methods of following metabolism of urediospores suggested that the control of urediospore germination perhaps depends on factors other than self-inhibitors or water balance. These studies emphasized the necessity for exactness in defining and providing pregermination conditions employed in studies of spore germination. Urediospores of <u>Puccinia graminis</u> were uniformly labeled with ¹⁴C to study the utlization of endogenous substrates at frequent intervals with small amounts of spores under conditions conducive to germination. Subsequent analyses indicated that the germination process in spores of obligate parasites is not based solely on the utilization of lipids. Some possible roles of the changes in internal and external pools of soluble carbohydrates are indicated.

B. Control of Virus Diseases

- 1. Vector of soil-borne mosaic clarified. Resting spores of the fungus Polymyxa graminis that had been exposed to 0.1N acid or alkali; germinated to give zoospores which transmitted the virus to wheat seedlings. All stages in the life cycle of the Nebraska isolate of this fungus were determined. Thus its identification was confirmed. Acid and alkali treatments, similar to those to which the zoospores were exposed, destroyed the partly purified virus as well as the virus in leaf extracts. From these reactions it was concluded that the virus is inside the resting spore of the fungus or in some way is protected by it.
- 2. Movement of wheat mosaic virus in plants. A 4-day dark treatment after inoculation with the wheat mosaic virus markedly increased the number of plants showing systemic symptoms in Nebraska studies. Presumably the virus remained localized in the roots of these plants until it moved up with reserve foods after the dark treatment. Removal of the plant tops caused a similar effect. Preliminary results have suggested that other cereal viruses tend to remain in the roots at low temperatures.

- 3. Labeled antibody technique applied. To trace the location of viral antigen at a subcellular level radioiodinated antibodies to viral protein were used in Nebraska. Hydrated sections of plant tissue are treated with specific antibodies labeled with I-125. After removal of the excess antibody I-125 the sections were covered with liquid emulsion and exposed in the dark for varying length of time, depending on the expected amount of antigen present in the sections. After fixation of the autoradiogram the presence of viral antigen was indicated by the appearance of exposed silver grains overlying the cell organs with which the viral protein is associated. Procedures have been found that satisfactorily immobilize tobacco mosaic virus (TMV) and soil-borne wheat mosaic virus (SBWMV). These virus are not washed out of opened cells, nor translocated, upon fixative removal if the tissue is infiltrated with gelatin in the presence of the fixative. The labeled antibody technique is extremely sensitive to small amounts of antigen. It is, thus, important to have as few non-specific antibodies present as possible in order to minimize the general background. The development of this technique for use with viruses is important in virus studies.
- 4. Barley yellow dwarf virus in wheat. The lower leaves of susceptible varieties of oats, barley, and wheat infected with barley yellow dwarf virus (BYDV) showed a marked loss of chlorophyll, reduction in photosynthesis, increase in dry weight, and generally an increase in respiration in South Dakota. However, the physiology of the diseased third leaf differed from the physiology of a diseased second leaf in both oats and wheat. In general, chlorophyll content was reduced in all tissues to the same degree while other physiological responses tended more toward the normal in the younger tissue of a diseased plant. Therefore, photosynthesis per unit chlorophyll may be well above normal in the upper leaves of a diseased plant. In oats and possibly also in wheat infected with BYDV, the respiration rate increased per unit fresh weight but the dry weight increased even more rapidly so that respiration per unit dry weight fell below normal. This differed from the pattern previously seen with barley.
- 5. Field trials with BYDV. Seven of 14 entries from the World Collection of Wheats, previously identified in a field screening trial as symtomless, were compared in replicated plots in South Dakota. Four of the seven yielded 89 to 98 percent of their virus-free control. The virus produced very slight effect on date of heading and only slightly more on height. Differences due to aphid numbers were not significant, but differences due to infection with BYDV, (whether 2, 4 or 8 aphids), were highly significant for heads per plant height, heading date, kernels per head, 1000-kernel weight, and weight of kernels per head in South Dakota tests (cooperative with Entomology Research Division). Highly significant differences due to plant age at time of infection were recorded for tillering, heads per plant, height and heading date. Varieties responded differently for these characters. In a field test to study the effect of BYDV on tillering, weekly inoculations were made over a span of 8 weeks. Beginning at one week after plant emergence, plot number 1 was inoculated; the second week after emergence plot number 2 was inoculated, and this pattern of inoculations continued until eight plots were inoculated,

one time each at 1 week intervals. A severe effect was produced on all yield components and many plant characters with the earliest infection. The effect of the virus on most characters steadily decreased with each later inoculation.

- C. Smut and Root Rot Studies in the Pacific Northwest Disease Laboratory
- 1. Calonectria nivalis, the perfect stage of <u>Fusarium nivale</u>. In 14 counties in Washington, Idaho, and Oregon, <u>C. nivalis</u> was found during 1967. Perithicia occurred in the basal leaf sheaths of winter wheat, barley, and rye, which were parasitized by <u>F. nivale</u> (a "mild" form of foot rot). It was not found on spring grains. Fertile perithecia capable of ejecting ascospores were found in the field from June to November. By December 1967, all perithecia examined were empty.
- 2. Saprophytic colonization of straw by <u>Fusarium culmorum</u>. F. culmorum colonized wheat straw buried in field soil in the laboratory at Pullman, Wash., but there is little evidence that such colonization occurs under field conditions. Therefore, it does not appear to be important in a build-up of this fungus in the field. When plants in the greenhouse growing in Palouse silt loam soil with 9,200 propagules of F. culmorum per g were subjected to a prolonged soil moisture stress, more premature blighted plants occurred than when plants grown in lots of the same soil were watered freely. There were three to four times more blighted plants in the drier soil.
- 3. Chlamydospore germination and the growth and survival of germlings of F. culmorum were studied in non-sterile soil. Percentage germination was high (35 to 50 percent in 20 hr.) in soil amended with either glucose or sucrose and inorganic nitrogen in the form of ammonium sulfate or potassium nitrate. Germination was poor (5 to 10 percent) when sugar alone was added and there was no germination when nitrogen or water alone were added. Germination occurred uniformly well when the water contents ranged from field capacity (FC) to below permanent wilting percentage (PWP) in both Ritzville fine sandy loam and Palouse silt loam soils. Germling growth and survival, on the other hand, was best only in soil at PWP or drier. In wetter soils, germlings lysed or were converted into new chlamydospores. When wetter soils were used the rate of germination and lysis were both greatest at 25°C. Germination and lysis both occurred in wetter soil at 10°C but at 4°C there was none. That lysis occurs in wet soil may account for the fact that F. culmorum causes its greatest damage to wheat in dry soil. In 24 hr., 20 to 30 percent of the chlamydospores germinated in soil near wheat seeds and 14 to 20 percent germinated near wheat straw. Lysis of the germlings occurred near seeds in soil near FC but not in soil near PWP. Germination was poor near seminal roots. The range was from 0 to 6 percent in 24 to 48 hr. This may be why F. culmorum is rarely isolated from seminal roots of plants collected from fields.

- 4. Collections of flag smut from the Pacific Northwest. Twenty-six flag smut collections evaluated for pathogenicity on 10 varieties of wheat were similar and they represented 1 pathogenic type of smut. None of the 26 collections were pathogenic to Golden, Norin 10/Brevor-14, or Dixon 115. It was apparent that the spread of flag smut in the Pacific Northwest was from spore dissemination by wind from the original point of detection in Klickitat County, Wash.
- 5. Oxathiins control seed-borne but not soil-borne bunt. DCMO (Vitavax*) controlled seed-borne common bunt at all rates of application to the seed, from 1 to 8 oz./bu. in Washington tests. In contrast, DCMOD (Plantvax*) controlled this seed-borne common bunt only at the highest rate of 8 oz./bu. Other formulations that contain hexachlorobenzene (HCB) and TCNA (-2,3,5,6-tetrachloronitroanisole) also controlled seed-borne common bunt in these tests. In addition, good control of soil-borne common bunt was provided by Vitavax at 2 to 8 ox./bu.

Plantvax and Vitavax failed to control dwarf bunt when applied to seed or used as foliar sprays in Montana.

- 6. Storage life of spores of Tilletia caries and T. foetida. Spores of 17 races of T. caries and T. foetida, stored in the laboratory in Washington as smut balls and as powdered spores, retained their viability for at least 22 years but had lost viability at 30 years. One race had lost viability in 18 years.
- 7. Self inhibitors present in common and dwarf bunt teliospores. Trimethylamine contributes to self inhibition. Four other volatile amines have also been detected.
- 8. Mechanism of light-induced sporulation in Ascochyta pisi. In Oregon the production of a sporogenic substance was involved in sporulation of A. pisi which was induced by light. The molecule of this substance is known to contain six hydroxyl groups, a single nitrogen atom and at least one, possibly more, sugar moieties. Nitrogen metabolism plays a role in the intensity of the sporulation response. Greater sporulation resulted when the fungus was supplied with rich sources of amino acids. The presence of either iron or zinc is necessary for photoinduction.
- 9. Resistance to bunt. In Washington, Columbia bunt resistance to T-5 and T-15 (Rio or Turkey gene) was shown to be associated with glume color on chromosome lB in a cross of Columbia x Elgin. The M₁ gene, which governs resistance to T-16 race of bunt segregated independently of the Turkey type resistance and glume color. A hybrid of race T-13 x L-8, has been isolated which is virulent on P.I. 166921. In prior studies P.I. 166921 exhibited resistance to all races of common and dwarf bunt. A second spore line has been isolated from T-5 which attacks the Bt-3(K) gene. It was isolated by passing T-5 through the Hohenheimer x Elgin differential, the same procedure used to isolate the first X-1 selection. The T-5 culture used in this study did not attack the Bt-3(K) gene carried by the P.I. 138383 x Elgin

differential. Thus, X-l does not appear to be a mixture in T-5.

10. Plant height and loss of yield due to foot rot not associated. At Pullman, Wash., the variety Burt is susceptible to Cercosporella foot rot whereas Nord expresses tolerance. When lines of various culm lengths within the populations Sel. 14/7*Burt and 14/5*Nord were grown under foot rot inoculated and uninoculated conditions, foot rot reduced height to a comparable degree within the highly susceptible populations (Burt background) and within the tolerant populations (Nord background). However, the culm length was not associated with losses caused by the foot rot.

RPA 207 - CONTROL OF INSECT PESTS

- 1. New virulent race of greenbug attacks winter wheat. Oklahoma developed a resistant variety Ponca, Kansas a resistant Bixon and Texas a resistant Concho, based on previously demonstrated resistance (DS28A) to greenbugs. (Cooperative with Entomology Research Division). A new field race of greenbug is vigorously and severely damaging these varieties. Hence, they have limited immediate use without combined resistance to the new race. Resistance has been noted in rye, oats, and barley but nothing thus far tested in wheat.
- 2. Cereal leaf beetle resistance makes progress. In Michigan and Indiana trials, high levels of resistance were noted in four common wheats and several tetraploids. Crosses between commercial varieties and resistant forms in Michigan, Indiana, North Dakota and South Dakota, give promise of considerable protection to both winter and spring wheats. Pubescence on leaves continues to be one mechanism for resistance but probably is not the only one. It has been determined in Michigan that a dense covering of hairs on the leaves retards feeding by the greenbug and possibly the laying of eggs.
- 3. Hessian fly resistance breeders take on new race forms. While forms or races have been noted nationally for many years, the problem has been relatively stable within regions until very recently. With four and possibly five races in the eastern States, new varieties are needed to combat this situation. Fortunately, Ribeiro resistance gives protection to all races and combinations of other variety genes.

RPA 405 - PRODUCTION OF FIELD CROPS WITH IMPROVED CONSUMER ACCEPTABILITY

A. High Lysine and High Protein

Lysine and protein analyses. At Lincoln, Nebr., the analyses of 3,500 lines from the World Collection and of 40 spring wheats with suspected high lysine or high protein properties indicated that sufficient genetic variability exists for successful selection of high lysine wheats at different protein levels. The highest lysine/protein content in the 3,500 lines was 4.2 percent and the mean was 3.01 percent. One percent of the lines contained more than 3.55 percent. Protein ranged from 9.0 to 21.1 percent (dry matter basis) with a mean of 13.4 percent. About 1 percent of the lines had greater than 17.4 percent protein.

2. Heritability and correlation among agronomic and quality characters. Four crosses between 2 soft red winter wheats, Atlas 50 and Atlas 66, and 2 hard red winter wheats, Triumph and Kaw, were studied for heritability and correlation values among various agronomic and quality characters at Manhattan, Kans. Flour yield, 1,000 kernel weight, and kernels per 30 ml were more heritable than test weight, protein content, and mixogram mixing time. The parent cultivars were separated by 2 to 4 factors for flour yield, 3 to 4 factors for protein content, and at least 2 factors for mixing time. Among all the combinations of 13 characters studied there were significant correlations between 7 pairs. Thirteen progenies of the crosses, Kaw x Atlas 50 and Atlas 50 x Kaw, contained 1 to 3 percent more flour protein than Kaw controls, and possessed promising hard wheat characters for milling, baking, and dough mixing properties.

B. Role of Flour Components

1. Water-soluble fraction of flour. It has been shown conclusively at Manhattan, Kans., that the water-soluble fraction of flour is not responsible for bread-making quality differences. However, water solubles are required to produce a normal loaf of bread. They were found to have a dual role by contributing to gassing power and by contributing to gas retention in modifying the physical properties of the gluten. The dialyzable fraction of the water solubles contribute to gas production and the gycoproteins are involved in the modification of the gluten. Baking quality is located in the gluten proteins.

From results of extramural research at Fargo, N. Dak., it was concluded that the water-soluble protein components of good quality amber durum wheats contribute to the desirable yellow color of spaghetti, while in poor quality amber durum varieties it appears to cause darkening of the color. Varieties which contained large amounts of basic proteins (Fraction A) were associated with off-color spaghetti. For evaluating spaghetti quality, objective methods of measuring color and cooked spaghetti tenderness were developed.

2. <u>Lipid fractions</u>. At Manhattan, Kans., when free polar lipids were added to petroleum-ether extracted flours, the loaf volume increased substantially; when bound polar lipids were added the increase was smaller. Total free lipids that contained a mixture of nonpolar and polar components in a ratio of 3:1, improved bread quality less than polar lipids alone. Nonpolar lipids decreased loaf volume and impaired crumb grain of bread. The effects on bread depended on levels and ratios of the nonpolar to polar lipids. Lipid fractions isolated from various flours indicated that there are no varietal differences which contribute to lipid actions.

Also, at Manhattan, total (unfractionated) and nonpolar wheat flour lipids substantially increased the length of time needed to reach the point of minimum mobility in the parinograph and in the mixograph. Water absorption was unaffected and mixing tolerance was improved by adding nonpolar or total free lipids. The baking strength of flour from which these lipids were extracted had no effect on the contribution of the lipids to mixing

characteristics. The effects of lipids on mixing increased with an increase in levels of lipids added. Nonpolar and total flour lipids had similar effects on mixing, whether added to untreated or to petroleum-ether-extracted flours. Temperature of peak hot paste viscosity was lowered about 4° by adding 2 percent flour lipids. Non polar lipids increased the peak viscosity; polar lipids had little effect.

C. Milling and Altered Behavior

- 1. Starch damage. By ball-milling flour and starch under closely controlled conditions in Wooster, Ohio, it was possible to predict the quantity of damaged starch that was generated by the process. The extent of starch damage in prime starch is linear with ball-milling time (within limits), but because of the protective nature of gluten and other components, starch damage proceeds much more slowly (and logarithmically) in flour. Progressive ball-milling action improved cake volume which coincided with the increase in the number of particles in the 20 to 30 micron range. Continued ball-milling was detrimental to cake quality, and resulted in high starch damage with little additional reduction in particle size.
- 2. Measuring starch. An enzymatic procedure for the determination of starch in wheat developed at Wooster, Ohio, has been found to be applicable to the estimation of the fraction with normal amylose:amylopectin ratio in corn. The test may be useful in evaluating the nutritive value of feeds containing corn (and wheat).
- 3. Influence of size of kernels and bleaching on milling. Wheat lots were sized according to kernel size at Fargo, N. Dak. The data show that the dollar value can be increased if a wheat lot is first sized, then milled and the flours blended after milling. There is a decrease in patent and total flour extraction as the percentage of small kernels is increased. Four varieties of wheat were artificially bleached to determine the effect of bleaching on the milling characteristics. The data show a significant improvement in the milling results after bleaching.

D. Flour Quality In Standard Products

1. The gluten protein factors. The factor or factors responsible for breadmaking quality differences continue to point to the gluten protein. At Manhattan, Kans., the ultracentrifugation of gluten solutions (0,005 N lactic acid) at 100,000 x G separated the proteins too large to enter the starch-gel (glutenins) from the proteins migrating into the starch-gel (all the gliadin plus 65 percent of the glutenins). Bread baked from extracted flour into which the two fractions were reconstituted had loaf volume and crumb grain equal to that of the original flour. By interchanging the two fractions from good and poor quality varieties it was shown that the proteins which migrated into the starch-gel were responsible for loaf volume potential. Further fractionation of the proteins which migrated into starch-gels by either untracentrifugation at higher R.C.F. (198,000 xG) or by precipitation

with 70 percent ethyl alcohol, yielded fractions which replaced the non-migrating proteins in baking.

- 2. Mixing quality. Mixogram tests were discontinued at Wooster, Ohio, on cake and cookie type wheats because of the relative insensitivity but AWRC (alcohol-water-retention capacity) tests continue to show good prediction value.
- 3. Classifying wheat types. Protein content plus a modified sedimentation test classified western wheats (Pullman, Wash.) into club, common soft, and common hard wheat types with low level of error.

-- Rye --

RPA 307 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY

A. Breeding and Genetics

- 1. Inbreeding to develop more useful ryes. A few inbred lines of rye have survived rigid selection and inbreeding at Tifton, Ga. They are from several States, Brazil, Kenya, Turkey, and Spain. Several hundred individual selections of Gator rye are maintained by inbreeding. These are propagated by self-pollination. They are resistant to leaf rust and mildew and they have good seed quality.
- 2. Amphiploidy. Under a P.L. 480 grant, workers in Spain doubled the chromosomes of five species, crosses, and several rye varieties with colchicine and returned them to this country. The five fertile amphiploids and nine tetraploid varieties of rye are under test at Tifton, Ga.
- 3. Chromosome substitution. A substitution of chromosome III of rye for chromosome 2D of wheat was obtained at Columbia, Mo., adding to the previously obtained substitutions of rye III for 2A and 2B. This rye chromosome belongs to homoeologous group 2 and should be designated 2R.
- 4. Triticales tested show deficiencies. The triticale nursery at St. Paul, Minn., was made up of six experimentals (wheat x rye), two spring wheats (Manitou and Pitic 62), one oat (Harmon), and one barley (Conquest) variety. None of the triticales yielded as well as the two wheat varieties.
- 5. Limited numbers of triticale trials grown. Most of the triticales grown in nursery trials at College Station and Denton, Tex., were from Canada. None was impressive in performance. Most were approximately 50 percent sterile, tall, lacked winterhardiness, and were disease susceptible. Fifteen winter type triticales were obtained from Canada. These were increased in the greenhouse at Lincoln, Nebr., in 1966 and grown in the field at Lincoln. Survival ranged from 0 to 95 percent. Two that survived will be used in a triticale breeding program. Yields from the best one in which 95 percent of the plants survived were only two-thirds that of the Gage variety plot with which it was paired. At Stillwater, Okla., 27 strains of triticale were

Data obtained from these entries included culm diameter, plant height, spike length, and seed set. All characters exhibited a high degree of variation, and sterility was relatively high. The average seed set was 47 percent.

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-- Wheat --

RPA 307 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY

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OAT AND BUCKWHEAT BREEDING, GENETICS, DISEASES, QUALITY, AND PHYSIOLOGY

Crops Research Division, ARS

USDA and Cooperative Program

	: Scientist Man-Years F.Y. 1968									
Location of Intramural Work	: Research Problem Area :									Total
	:	307	:	208	:	207	:	405	:	
	:		:		:		:		:	
Oats	:		•		:		:		:	
Maryland (Beltsville)		_		1.2	:	0.1	:	0.7	:	2.0
Florida	:	-	:	1.0	*	-	:	-	:	1.0
Idaho	:	0.3	:	-	:	-	:	-	:	0.3
Illinois	:	-	:	1.0		-	:	-		1.0
Iowa	:	0.8	:	1.2	:		:	-	:	2.0
Michigan	•	-	:	-	:	0.5	:	-	:	0.5
Minnesota	:	0.2	:	1.0	:	-	:	-	:	1.2
Mississippi	:	-	:	0.1	:	-	:	-	:	0.1
New York	•	-	:	1.0	:	-	:	-	:	1.0
Pennsylvania	:	0.9		-	:	-		-	:	0.9
South Dakota	:	-	:	-		0.6	:	-	:	0.6
Texas		0.1	:	0.2	:	~	:	-	:	0.3
Washington	:	-	:	0.3	:	-	:	-	:	0.3
Total Oats	c.	2.3	•	7.0	9	1.2		0.7	9	11.2
	0		0						:	
Buckwheat	0				9					
Pennsylvania		0.1	0 0	2000		_	0	-	0	0.1.
Total Buckwheat	0	0.1	0	-	:	-	e 0	-	:	0.1
Total	0	2.4	6	7.0	41 0	1.2		0.7		11.3

Intramural program is supplemented by extramural support representing P.L. 480 funds in 1 country representing 67,600 U.S. dollars equivalent.

Problems and Objectives

Oats are grown in the U.S. mostly for feed; some of the crop is processed for human consumption. There has been a decline in the amount of oats planted since 1962 because of the lack of stability of production mostly due to pests. The 21 million acres planted in 1968 are 3 percent above the 1967 plantings but this amount remains 19 percent below the 10-year average. Generally, the crop is produced under warm, humid conditions which favor the development of disease and insect pests, and in some years, major losses still result from crown and stem rusts and barley yellow dwarf virus. There is a pressing need for more disease-resistant varieties. Additional sources of winterhardiness and information on effect of diseases on winterhardiness are also needed. Improvements in yield and nutritive quality would make oats more valuable as food and feed and as a desirable grain crop for production.

Buckwheat is grown on limited acreage and has received minor attention. Major improvements in yield and quality are needed to make it a competitive feed for livestock, its primary use.

Major objectives of research are:

- 1. Develop high yield, stiff-straw; disease resistant high protein oats.
- 2. Discover and utilize new sources of resistance to diseases and insects.
- 3. Develop oat varieties with improved nutritional quality.
- 4. Determine inheritance of resistance to disease and insect pests.
- 5. Control diseases and insects by cultural, chemical, and other safe effective means.

Progress - USDA and Cooperative Program

-- Oats --

RPA 307 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY

A. Breeding and Genetics

1. New varieties developed. Norwin (C.I. 8018) was developed and released cooperatively with the Texas Agricultural Experiment Station for Northcentral Texas and the Rolling and High Plains areas. It is equal to or ranks higher than varieties presently grown in these areas.

Pettis, an early spring oat, was released with Missouri Agricultural Experiment Station. It is suitable for States of the North Central Region. Seeds have a high test weight, thin hulls, and high groat percentage. It is valuable for its high tolerance to barley yellow dwarf virus.

Also released with the Texas Agricultural Experiment Station, and for southern Texas and the Gulf Coast was Coronado (C.I. 8260). It is superior in its resistance to crown rust and has good yielding ability, superior straw strength, and plump and attractive kernels.

A medium early oat variety, Pennlan (C.I. 7881), was developed in a Federal-State program and released with Pennsylvania. It is suited for production in southcentral and southeastern Pennsylvania and areas of similar climate. Its principle assets are short straw and early maturity.

Two oat multilines, Multiline M68 (C.I. 8346) and Multiline E68 (C.I. 8345), were developed in a Federal-State program at Ames, Iowa, and released with the Iowa Agricultural Experiment Station. They are especially valuable to provide longer lasting resistance to crown rust and to delay the onset of epidemics. These lines are a result of a new concept in oat breeding which promises better crown rust resistance.

- 2. New Composite Cross germ plasm. Composite Cross II was released to breeders for germ plasm with California Agricultural Experiment Station. This composite is made up of the cultivated type segregates from the cultivars x Avena fatua that have been made in the Federal-State program since 1947. The A. fatua parent has contributed early maturity, shatter resistance and both specific and nonspecific resistance to the important oat diseases. Composite Cross II should be particularly valuable for developing oat cultivars for desert areas and for areas where very early maturity is desired.
- 3. New hexaploid and tetraploid species of oats. At Beltsville, Md., apparently new and valuable sources of various levels of resistance to crown rust, stem rust, barley yellow dwarf virus, soil-borne oat mosaic, smut, mildew, and probably other oat diseases have been found among collections of the hexaploid Avena sterilis from Israel and other Mediterranean countries. Unusually high protein content of groats and high groat weight have been found among certain lines of A. sterilis and in the new tetraploid species A. magna. In general, these characters will need to be combined in adapted varieties through hybridization.
- 4. Chromosome associations of Avena magna. The Avena spp. constitute a polyploid series with 2n=14, 28, and 42 chromosomes. In the recently discovered tetraploid A. magna, karyotype analysis and meiotic studies made at Ames, Iowa, suggest that a genome in A. magna is partially homologous to the one in the diploid species, A. strigosa, and to one in the tetraploid species A. abyssinica. The A genome is the only one that can be partially homologous in all three species. A. magna appears to be closely related to the hexaploids morphologically and in chromosome association in pentaploid hybrids. There were indications that A. magna may have two genomes partially in common with two genomes in the hexaploid species (A. sterilis), a common genome and the remaining genomes partially common, or two genomes partially in common with the three genomes of the hexaploid. This new tetraploid

- species, A. magna, that has close morphological resemblence to the hexaploid species, A. sterilis, is of interest because it may prove to be useful in elucidating the origin of hexaploid oats.
- 5. Chemical mutations for crown rust resistance. Mutations of oats from seed treated with ethyl methane-sulfonate (EMS) at Ames, Iowa, expressed quantitative resistance to crown rust. A very few lines superior for resistance were identified, however, the fact that mutations produced by EMS showed differential responses to crown rust points up the fact that reactions to crown rust can be modified by chemical mutagens. X-ray mutations have not shown the same reaction.
- 6. Germ plasm bank. During the year, more than 5,000 accessions were added to the World Oat Collection from P.L. 480 research in Israel. They originated from seven foreign countries and contained species such as Avena sterilis, A. barbata, A. wiestii, and A. longiglumis.
- 7. Identification of monosomics. Progress was made at Ames, Iowa, in establishing and identifying monosomic lines. Karyograms of all monosomics are being prepared as an aid in identification. Through karyotype studies, monosomics have been identified for six chromosomes of the A's genome and for nine of those from C and D genomes.
- 8. Monosomic analysis. Monosomic lines have been used to determine that the gene for crown rust resistance derived from the diploid, Avena strigosa is not located on chromosomes 7, 14, or 21. Monosomics were used also to determine that the A gene for resistance to race 7A of stem rust is not located on chromosomes 2, 5, 6, or 21. These analyses were made at Ames, Iowa.

RPA 208 - CONTROL OF DISEASES

- 1. Late rusting in oats. Red Rustproof oats has shown a late rusting character that can be transferred to an early maturing variety with good agronomic characters. In Florida, two crosses with this variety produced some progenies which exhibited late rusting. This late rusting is only expressed in the field and investigations are being made to determine what conditions it.
- 2. Resistance to Helminthosproium avenae. In the Southeastern United States, H. avenae causes the most destructive disease. It produces both leaf blight and black stem. Among promising selections from the Quincy, Fla., nursery, three were found that have resistance to both phases of the disease. These three lines are being incorporated into oat varieties that are resistant to H. avenae. Observations and tests have indicated that pathogenic strains of the fungus are prevalent in Florida.
- 3. Victorin toxin activity. In studies in Florida on the toxin, victorin, it was observed that cytokinins (a group of compounds that stimulate protein synthesis) appeared to increase the sensitivity of susceptible tissue to the

toxin. However, it was later determined that cytokinins neither influenced sensitivity of the host nor the toxicity of the toxin. It caused the tissue to absorb more toxin. A wide variety of cytokinins increase toxin absorption, and they may influence the effectiveness of wilt producing pathogens under field conditions. It was observed that all of the cytokinins that increased toxin absorption were aminopurines. The results of tests lead to the conclusion that the plasma membrane is the site of action of victorin, thus it should be possible to determine how wilt toxins kill plant cells.

- 4. Purifying victorin. In purifying victorin at Gainesville, Fla., the final substance from the biogel column was 60 times more pure than the culture filtrate. Further improvements will have to be made to increase the amount so as to have enough purified toxin for elemental determinations and spectral analysis.
- 5. Light rusting or tolerance to stem rust. Selections made in Puerto Rico in 1967 because of their light rusting or tolerance in 6AFH nursery were retested in Minnesota. Some were found to have hypersensitive resistance to race 6AF. At least four entries out of 39 total selections tested showed a marked reduction in numbers of infections and development of pustules. This characteristic may be important in affecting rates of increase of stem rust epidemics. These oat species came from a P.L. 480 research project with Israel.
- 6. Crown rust resistance from Avena sterilis. The Avena sterilis backcross program to incorporate crown rust resistance from A. sterilis into adapted cultivated oats was started in Texas. Some of the F_1 's did not carry the sterilis gene for resistance. This may have been due to some heterogeneity in some of the sterilis population used as parents. Recently, improved seedling testing methods have made it possible to select resistant segregates from the backcross progeny easier than before.

In Iowa two A. sterilis strains that were resistant to crown rust were morphologically different, but they carried the same gene or genes for resistance. Two other equally resistant strains apparently carried distinct alleles of the same gene at another locus. It was also shown in tests in Iowa that dominance and degree of resistance afforded by various genes varies considerably with relatively small environmental differences.

In special nurseries of A. sterilis in Texas, many of the strains had excellent resistance for crown rust, but very few showed promise as sources of resistance to stem rust.

7. Barley yellow dwarf virus (BYDV) and physiology of Clinton oats seedlings. There is a sharp decrease in rate of photosynthesis in diseased leaves. Generally the chlorophyll content in diseased materials decreases by 40 to 45 percent while the dry weight increases by a similar amount. However in experiments with Clinton oats at Brookings, S. Dak., the photosynthesis was reduced by about 65 percent in both the second and third leaves. Thus,

photosynthesis per unit chlorophyll was well below the normal. The stimulation of respiration caused by BYDV found in oats is about the same as that in barley and less than that in wheat.

- 8. Purifying and assaying the RMV type of BYDV. It became desirable to purify and assay the RMV type of BYDV transmitted specifically by Rhopalosiphum maidis when the serological relationships became known among RPV. MAV. and PAV types of BYDV, transmitted most efficiently by Rhopalosiphum padi, Macrosiphum avenae, and pea aphid, respectively. The RPV, MAV, and PAV had already been purified. Eventually, relationships among all four of the major types of BYDV will be determined. In New York. injection assays of the partially purified RMV showed that the preparation was infectious. RMV could also be assayed by the membrane-feeding procedure. R. maidis fed readily through the stretched paraffin membrane. These experiments showed that both bioassay procedures can be applied to this RMV isolate of BYDV. They also showed that three types of BYDV are similar in regard to the disease they cause, host range, persistence in their aphid vectors, morphology, and sedimentation of the infectious particle. However, RPV appears to be unrelated to MAV and PAV on the basis of serology and interaction of virus isolates in plants. The relationship of RMV to the other virus types has not been determined. These observations on relationship suggest a range of variation for BYDV that may be found to overlap with variation among other groups of viruses. Thus the evolution of plant viruses is not necessarily restricted and narrow.
- 9. Loss of vector specificity for BYDV isolates. Loss of specificity occurred regularly in tests on oats inoculated simultaneously with the two virus isolates, MAV and RPV at Ithaca, N. Y. Following double infection with these two types of BYDV, R. padi, usually transmits the virus that is subsequently transmissible only by M. avenae. The loss of specificity occurred only when R. padi was exposed to MAV before being exposed to RPV. This is not a frequent phenomenon but it is consistent so that its occurrence must be considered in attempts to explain the mechanism of vector specificity.

RPA 405 - PRODUCTION OF FIELD CROPS WITH IMPROVED CONSUMER ACCEPTABILITY

1. Groat protein content. A relatively high protein line of Avena sterilis was crossed with five other oat varieties at Beltsville, Md. There was a strong expression of heterosis for groat weight in F_1 's and transgressive segregation for groat protein content and groat weight in the F_2 generations. Distributions in the F_2 populations indicated that the majority of genes for high protein were possessed by the A. sterilis parents. Preliminary tests of the crosses studied indicate that the oat groat protein content is controlled by multiple genes. Environment is also a major factor in oat groat content.

-- Buckwheat --

RPA 307 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY

A. Breeding and Genetics

- 1. Pennquad. This first buckwheat variety released from an Experiment Station, has attracted quite a bit of attention since its release cooperatively with Pennsylvania in 1967. It's yields remain good, and preliminary reports have indicated that its milling characteristics are satisfactory.
- 2. Autotetraploid breeding. In attempts to isolate a tetraploid form with somewhat smaller and plumper seed than that of Pennquad, F4 plants from a Pennquad x autotetraploid La Harpe cross were examined. Selections were isolated which have plumper seed, large stiff stems, outstanding lodging resistance, and improved fertility over La Harpe.
- 3. Genetic studies on stylar length. In genetic studies using 1,336 F_2 plants from 9 crosses, it was determined that at least 3 genes appear to be involved in stylar length. Because of a pollen inhilition system in long styles, it is believed that self-fertility could be expected where the stylar length is reduced one-third or more.

Publications - USDA and Cooperative Program

-- Oats --

RPA 307 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY

Breeding and Genetics

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RPA 208 - CONTROL OF DISEASES

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-- Buckwheat --

RPA 307 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY

Breeding and Genetics

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RICE BREEDING, GENETICS, DISEASES, QUALITY, CULTURE AND PHYSIOLOGY

Crops Research Division, ARS

USDA and Cooperative Program

	: Scientist Man-Years F.Y. 1968									
Location of Intramural Work	:		:	Total						
	:	307	•	208	:	207		405		
					:		:		:	
Rice	:		:						:	
Maryland (Beltsville)		1.3		0.2	:	-	:	-	:	1.5
Arkansas		0.8	:	0.2	:	_	*	-		1.0
California		1.0	:	-	:	-		-	:	1.0
Louisiana (Baton Rouge)		-	:	0.4	:	-		_		0.4
Louisiana (Crowley)	:	0.8		0.2	:	-		-	:	1.0
Texas	•	0.9	:	0.9	:	_		1.2	:	3.0
Total	:	4.8	:	1.9	•	-	:	1.2	:	7.9

Intramural program is supplemented by P.L. 480 funds in 2 countries representing 50,700 U.S. dollars equivalent.

Problems and Objectives

Rice is produced under many environmental conditions and it is exposed to many diseases and pests. Consumers and processors require diverse types. Rice research must cover a wide variety of problems in order to develop varieties and cultural practices that will give maximum production of the desired types of rice. High yielding varieties are needed that resist lodging, diseases and insects; respond to high rates of fertilizer and to favorable cultural practices; have high nutritive value; and have desired cooking, processing and milling characteristics. In order to attain these desired types of rice, breeding for improvement, disease control, cultural practices, and quality evaluation must be coordinated.

Major objectives of the research are to:

- 1. Determine the genetics of growth, reactions to pests, and grain characters.
- 2. Develop disease control methods.
- 3. Identify factors affecting nutrition and processing of rice.
- 4. Develop cultural practices that give maximum rice yields.
- 5. Use genetic and other information to develop improved varieties.

Progress - USDA and Cooperative Program

-- Rice --

RPA 307 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY

A. Breeding and Genetics

- 1. Genetics of amylose content of the grain. Amylose content of the kernel is closely associated with the cooking and processing characteristics of rice. The mode of inheritance of amylose content was studied in two crosses at Beaumont, Tex. In a case where the amylose content of the parents differed as much as 7 percent, amylose content was controlled by one pair of genes of major effect and a few additional pairs of modifying genes. In a case where the parents differed only 3.5 percent, amylose content was controlled by a few pairs of modifying genes of small and approximately equal effect.
- 2. <u>Introduction of rice varieties</u>. There were 351 rice varieties from 10 countries introduced and grown in detention at Beltsville, Md. About 200 new introductions and about 260 World Collection varieties not previously classified were grown at Stuttgart, Ark., and Beaumont, Tex. The new introductions include varieties recently developed and released in other countries.

- 3. Cold water tolerance. At Beltsville, Md., a method was developed for evaluating tolerance of rice plants to cold water. The seedlings are grown for 26 days in fertilized soil which is submerged in about 15 cm of water at 15.5 C. Seedling height at end of the growth period is the criterion of cold water tolerance.
- 4. Morphological characters associated with straw strength. Studies on the genetics of morphological characters associated with straw strength were conducted under a P.L. 480 research project in Taiwan. The characters studied were the degree of extrusion of the panicle from the sheath, breaking strength of the straw and plant height. Each characteristic affecting lodging resistance was a quantitative character and the mode of inheritance was complex.
- 5. Evaluation of rice varieties in the World Collection. About 4,400 rice varieties from 57 countries from plantings in Arkansas, Louisiana, Texas, and Mississippi were evaluated. The length of growing period, kernel size and shape, and several physical and chemical endosperm characteristics associated with rice cooking and processing behavior were determined. Days from seeding to heading were not highly correlated with the other characters studied. Correlation coefficients between kernel length, kernel width, and length-width ratio were significant. Of the characters studied, amylose content, starch-iodine-blue values, and alkali reaction values were the best indicators for predicting parboiling-canning stability. For long grain U.S. varieties, protein content also was found to be an important component for influencing the parboiling-canning stability.

RPA 208 - CONTROL OF DISEASES

A. Evaluating Varietal Reaction to Pathogens

- 1. Varietal reaction to the kernel smut pathogen. Kernel smut is caused by Tilletia barclayana. Infection occurs at flowering and so rice must be grown to maturity before reaction to this fungus can be determined. In Arkansas, experiments showed that when nitrogen fertilizer rate is low and flowering occurs in mid-August kernel smut infection is significantly lower than when nitrogen rate is high and flowering occurs in mid-September. Late seeding in soil with excessively high nitrogen levels proved to be satisfactory in screening varieties and breeding lines for kernel smut resistance.
- 2. Testing for blast resistance. Rice varieties and breeding lines are tested for reaction to <u>Piricularia oryzae</u> in upland nursery beds in Texas. Moisture conditions favorable for leaf infection are provided by a sprinkler system. Seedling plants are tested in small closely spaced rows. This makes possible the testing of several plantings per year so that large numbers of breeding lines can be evaluated each year.

3. Genetics of reaction to <u>Piricularia oryzae</u>. The mode of inheritance of reaction to four races of <u>P. oryzae</u> was studied under a P.L. 480 project in Taiwan. F_2 populations and F_3 lines from crosses among 28 japonica rice varieties were evaluated. Reaction to each race was controlled by a single dominant independent gene. None of these four genes was linked with nine marker genes for other characteristics that were involved in these crosses.

RPA 405 - PRODUCTION OF FIELD CROPS WITH IMPROVED CONSUMER ACCEPTABILITY

A. Evaluation of Processing Characteristics

1. Cookability test for specific quality characteristics. A laboratory cookability test to determine the suitability of rice breeding lines for specific brewing processes has been developed at Beaumont, Tex.

Publications - USDA and Cooperative Program

-- Rice --

RPA 307 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY

Breeding and Genetics

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ALFALFA BREEDING, GENETICS, DISEASES, PHYSIOLOGY AND BIOCHEMISTRY Crops Research Division, ARS

USDA and Cooperative Program

	:	Sc	ienti	st Man-	Years	F.Y.	1968	
Location of Intramural Work		I	Total					
		307	:	208	:	207	:	
	:		:		:		:	
Alfalfa	:		:		:		:	
Maryland (Beltsville)		1.0	:		:	1.0	:	2.0
Kansas	:	0.7	:	0.1	:	0.2	:	1.0
Minnesota	:	1.9	:	0.1	:		:	2.0
Mississippi	:		:	0.4	:		:	0.4
Nebraska	:	0.4	:	0.4	:	0.2	:	1.0
Nevada	:	0.6	:	1.0	:	0.4	:	2.0
Raleigh, N. C.	:	0.6	:	0.7	:	0.7	:	2.0
Pennsylvania		0.3	:	0.9	:	0.4	:	1.6
Utah	:	0.3	:	0.1	:	0.1	:	0.5
Total	:	5.8	:	3.7	:	3.0	:	12.5

Intramural program is supplemented by extramural support representing (a) 1.1 SMY's at State Agricultural Experiment Stations 1, and (b) .8 SMY's at other U.S. institutions 2.

2/ RPA 207

^{1/} RPA 207 (.2 SMY) and RPA 208 (.9 SMY)

Problems and Objectives

Alfalfa and alfalfa-grass mixtures grown for hay in the United States occupy about 30,000,000 acres and provide more than 50% of the total hay required by livestock. The competitive position of alfalfa is in jeopardy, however, because advances in technology have not kept pace with most major crop plants. A substantial portion of the research effort has been necessarily devoted to solving emergency production problems such as developing resistance to spotted alfalfa aphids, pea aphids, alfalfa weevil, nematodes, and diseases attacking the foliage, root, and crown. Recent advances made in development of multiple disease and insect resistance, cytoplasmic male sterility, and basic information on expression of hybrid vigor in autotetraploids offer tremendous potential for increasing returns from alfalfa.

Major objectives of the research are to:

- (1) Develop alfalfa with increased potential for higher forage yield and improved quality.
- (2) Develop alfalfa with better disease and nematode resistance including better methods to reduce losses.
- (3) Reduce losses in alfalfa from insects through host plant resistance.

Progress - USDA and Cooperative Program

RPA 307 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY OF FIELD CROPS

A. Breeding and Genetics

- 1. Hybrid alfalfa. Significant progress was made as follows: (1) extension of genetic theory for autotetraploids, (2) development of inbred lines (20 lines from 2 germ plasm pools now in S4) to determine effects of inbreeding on hybrid and synthetic performance, at University Park, Pa., and (3) development of cytoplasmic male sterile lines and theory at Logan, Utah, and St. Paul, Minn.
- 2. Cytoplasmic male sterile lines developed. Plants with non-dehiscent anthers containing a limited amount of stainable pollen were isolated from 'DuPuits' alfalfa at Logan, Utah. Some of them yielded only sterile F_1 's and nearly sterile backcrosses when crossed to type "0" male fertile plants. These cytoplasmic male sterile plants were highly self-sterile but some pollen was functional in crosses with a recessive marker line. Female fertility was normal. Isolation of high combining lines and improvement of seed set are major objectives that need to be achieved in order to place production of commercial alfalfa hybrids on a firm economic basis.
- 3. Potential for increasing photosynthetic efficiency of alfalfa. Studies of factors affecting alfalfa leaf density and photosynthetic capability at

Beltsville, Md., demonstrated that natural genetic variation occurs in the crop for both characteristics.

- 4. Natural crossing in alfalfa increases with inbreeding. In Nebraska, percent natural crossing increased 24% from the S_0 to S_3 generations of white-flowered lines. Some Syn-1 synthetics from selected clones of S_1 and S_2 origin had 88-97% crossing as a 2-year average, a significant increase.
- 5. <u>Single vs. multiple row experimental plots</u>. Single row plots did not compare favorably with multiple row plots at Reno, Nev. The relationship between the two methods diminished rapidly after the second harvest year.

B. Quality

1. Resistance to insect pests improves quality. At Lincoln, Nebr., carotene contents of alfalfa synthetics resistant to spotted alfalfa and pea aphids were higher than those of susceptible entries when infested. Spotted alfalfa aphids reduced the protein content of susceptible synthetics, whereas protein content of resistant entries was unaffected. Mean percent reductions in forage yield, protein, and carotene contents were less for clones classified as resistant to potato leafhopper than those which were intermediate or susceptible.

RPA 208 - CONTROL OF DISEASES OF FIELD CROPS

- 1. Nonsegregating lines resistant to root-knot nematodes. A total of 25 winterhardy clones with test cross ratios indicating triplex or quadruplex genotypes with respect to Meloidogyne hapla resistance have been isolated at Reno, Nev. Test crosses have also been made on over 200 nonhardy clones to isolate resistance factors in triplex or quadruplex condition. The objective is to isolate nonsegregating lines for resistance to M. hapla, which in addition are resistant to other pests.
- 2. New hope for solving late summer unproductiveness. A breakthrough appears to have been made in solving late summer unthriftiness of alfalfa in the East. A field test of experimental lines in Maryland uncovered sizable differences in resistance to crown and root rot caused by Colletotrichum trifolii. Symptoms of the disease were like those commonly associated with late summer production slump, especially when moisture supply is ample. Disease inoculation techniques were adapted for testing large numbers of alfalfa plants for resistance and highly resistant plants have been isolated.
- 3. Biosynthesis of disease induced coumestrol. The time course of coumestrol accumulation in leaves inoculated with Ascochyta imperfecta at Raleigh, N. C., approximated the growth curve of the organism in culture. Coumestrol accumulation was not induced by non-pathogenic Ascochytas, Helminthosporium, or by wounding alone, but was induced by H. carbonum on wounded leaves. No incorporation into coumestrol of labeled cinnamic acid, acetate, glucose, or phenylalanine fed through stems was detected.

- 4. Taxonomy of Rhizoctonia solani. Four anastomosis groups in R. solani were demonstrated at Raleigh, N. C. Whenever isolates were paired on water agar, anastomosis occurred only between isolates within a group, but never between isolates of different groups. Of the 118 isolates studied, 46, 32, 21, and 10 isolates were assigned, respectively, to the 4 groups. Each group included isolates from several locations (at least two countries).
- 5. Calcium in middle lamella as related to stem nematode resistance in alfalfa. At Raleigh, N. C., calcium content of plant tissues was proportional to calcium concentration of the nutrient solution in which plants were grown and increased with age of plants. Calcium uptake by resistant plants did not play a central role in resistance, but reproduction of the nematode and disease reaction in susceptible plants may be influenced by calcium supply. This suggests that pectic enzymes may be important in phases of pathogenesis involving enlargement and separation of parenchymatous cells. In resistant plants, where this reaction is not successfully initiated, other factors affecting hyperplastic growth may be more important than calcium content of the tissues.
- 6. Heat treated clones remain free of alfalfa mosaic virus. Alfalfa plants infected with several alfalfa mosaic virus isolates were subjected to a continuous temperature of 35-36 C at St. Paul, Minn. Cuttings from treated plants were periodically assayed for virus infection. No recurrence of virus has been detected in cuttings ranging in age from 10 to 18 months from heat treated plants.
- 7. Mechanical vs. pea aphid transmission of alfalfa mosaic virus in alfalfa. Each of four virus isolates were transmitted to alfalfa clones mechanically and with pea aphids at St. Paul, Minn. A fifth isolate was transmitted to one clone by aphids only.
- 8. Culture media for Pseudopeziza medicaginis. Modified commercial oatmeal agar supported satisfactory growth and sporulation of \underline{P} . medicaginis at University Park, Pa. Media are easily prepared, and increased selectivity through the use of antibiotic amendments is promising.

RPA 207 - CONTROL OF INSECT PESTS OF FIELD CROPS

1. New pea aphid resistant alfalfas prove their worth. In much of Kansas and part of Nebraska, pea aphids destroyed most of the first crop of susceptible alfalfa varieties in the absence of insecticides. 'Dawson,' a resistant variety released in 1967 from the cooperative program with Nebraska Station, withstood damage from the pea aphid. 'KS-12,' an experimental synthetic in the Kansas program, had no symptoms of damage. KS-12 also has high resistance to the spotted alfalfa aphid and bacterial wilt, and in addition is an improvement over 'Cody' and 'Buffalo' in leafspot resistance.

- 2. Alfalfa germ plasm released for developing resistance to the alfalfa weevil. 'AWPX3,' which has moderate levels of resistance in laboratory tests for larval development, adult feeding, and forced oviposition, was released to alfalfa breeders. Field testing to select for adaptation, agronomic characteristics and tolerance mechanisms within this population will be necessary to utilize germ plasm effectively. Breeding resistance to the alfalfa weevil continues to be one of the most difficult and challenging objectives in alfalfa breeding. This is so because higher levels of resistance are needed and because plants and weevils each interact differentially with changes in environment. Development, seed increase, and field testing were accomplished at Beltsville, Md., Reno, Nev., and Raleigh, N. C., respectively.
- 3. Nature of alfalfa resistance to pea aphid. In Kansas studies, total nymphal days was a more reliable measure of resistance than total adult days, fecundity, or percentage nymphal mortality. Each of the criteria was adequate, however, to isolate highly resistant alfalfa plants. For most clones, excised branches were less resistant than intact branches and clone x intact-excised interaction was significant. Excised stems should thus be used cautiously in resistance studies.
- 4. <u>Saponins isolated from alfalfa varieties</u>. In extramural research in North Carolina, saponin mixtures from DuPuits and 'Lahontan' alfalfa were separated into individual components. This was followed by acid hydrolysis of individual saponins in order to identify the aglycone and sugar fractions. The components are being isolated and identified in order to determine their biological effects on alfalfa pests.

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CLOVER AND OTHER LEGUMES BREEDING AND GENETICS, DISEASES, QUALITY AND VARIETY EVALUATION, AND PHYSIOLOGY AND BIOCHEMISTRY

Crops Research Division, ARS

USDA and Cooperative Program

	: Scientist Man-Years F.Y. 1968							
Location of Intramural Work	:	Area	:	Total				
	:	207	:	208	:	307	:	
Sweetclover	:				*			
Nebraska	:	0.2			:	0.8	:	1.0
	:	0.2	:		:	0.0	:	1.0
Annual Clovers Mississippi (Stopoville)	*			0.2	:			0.2
Mississippi (Stoneville)	:		:	0.2	:	0 0	:	
Mississippi (State College)				0.1	:	0.9	:	1.0
Perennial Clovers			:			0.7	:	0.7
Colorado	*		:			0.7	:	0.7
Red Clover	:				:	1 0	:	1 0
Maryland (Beltsville)	:		•			1.0	:	1.0
Kentucky				0 5		1.0	:	1.0
Pennsylvania	:	0 =		0.5			:	0.5
Wisconsin	:	0.1		0.2		0.7	:	1.0
White Clover			•		:		:	
South Carolina				0.4		1.6		2.0
Total Clovers		0.3		1.4	:	6.7		8.4
Trefoil	:		:		:		:	
Maryland			:	0.5	:	0.5	:	1.0
Missouri			:	0.1		0.4	:	0.5
Mississippi	:		:	0.2		0.4	:	0.2
New York	:		:	0.2	:	0.8	:	0.8
Virginia	:		:	0.1		0.4		0.5
Crownvetch	:		:	0.1	:	0.4		0.5
Maryland	:		:	0.5	•	0.5		1.0
Colorado			•	0.5	:	0.3	:	0.3
North Carolina			:	0.1	•	0.3	•	0.5
				0.1		0.4		0.5
Missouri	•		•					
Virginia			•	0.1		0.4		0.5
Sericea Lespedeza			•	0 1		0 /		0 5
North Carolina			•	0.1		0.4		0.5
Lupines			•	0 0		0 5		0 7
Georgia (Tifton)			•	0.2		0.5		0.7
Desmodium sp.			•	0 0		0 5		0 0
Georgia (Tifton)	•		•	0.3	-:	0.5	•	0.8
Total Special Purpose Le	egumes	0.0	•	2.3	•	5.5	•	7.8
Total	•	0.3	•	3.7	•	12.2		16.2
<mark>Intramural program is suppl</mark> eme	ented b	y extr	amural	suppor	rt r	epresenti	ıng	(a) U.
SMY at State Agricultural Expe	eriment	Stati	ons $\frac{1}{\cdot}$,	and (b) P]	L 480 tui	nds	ın I
country representing \$15,021 l	J. S. dc	ollars	equiva	rent.	-	1/ RPA 2	207	0.3

Problems and Objectives

Clovers and special purpose legumes are utilized on more than 35 million acres of cultivated pasture and haylands; they are used on double this acreage as components of permanent pastures and rangelands and for soil and water conservation, roadbank stabilization, and wildlife management. At least 50 species are of regional or national importance. Species of major importance include red clover, white clover, crimson clover, sweetclovers, birdsfoot trefoil, lespedezas, lupines, and crownvetch. Especially needed are new varieties that are resistant to diseases and insects and that are fully adapted and most productive for a specific animal use in a specified environment. Improved plant performance may be achieved through known and newly developed methods of breeding, by interspecific hybridization, by newly developed methods of pest control or culture, or by newly discovered selection indexes of plant production efficiency.

The objectives of the research program with clovers and special purpose legumes include:

- 1. Development of new varieties and methods that improve quality and yield of forage.
- 2. Development of plant resistance and methods of control for diseases and nematodes.
- 3. Development of plant resistance to insect pests.

Progress - USDA and Cooperative Program

-- Clovers --

RPA 207 - CONTROL OF INSECT PESTS OF FIELD CROPS

1. Nature of resistance to sweetclover weevil. In contract research with the University of Nebraska, Deterrent B, found only in sweetclover weevil-resistant Melilotus infesta extracts, was isolated in crystalline form and sufficient material accumulated for structural determination. The white needle-shaped crystals have a melting point of 169 to 170°C, the compound sublimes at temperatures near the melting point at atmospheric pressure, is very soluble in water, soluble in methanol, slightly soluble in ethanol, and is insoluble in the more nonpolar solvents.

RPA 208 - CONTROL OF DISEASES OF FIELD CROPS

1. Persistence and forage yields of red clover increased by insecticide and insect-caging treatments. Plots of Pennscott red clover in Pennsylvania exhibited 82% survival 3-1/2 years after establishment when protected with insecticide soil drench and insect caging, as compared to 23% for check plots. The insecticide soil drench greatly reduced root damage by Sitona, but root rot was similar for all treatments.

2. Reduced growth of red clover associated with decrease in incidence and degree of internal breakdown (IB). A more frequent cutting treatment of red clover produced lower forage production, smaller root diameters, and lower incidence and degree of IB in Pennsylvania greenhouse studies. Mechanical restriction of lateral expansion of red clover crowns reduced incidence of IB. Red clover was grown under aseptic conditions 96 days before detection of contamination; IB developed in some plants of the treatment. All above studies support the earlier hypothesis that IB is a physiogenic disease.

RPA 307 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY OF FIELD CROPS

A. Breeding and Genetics

- Sweetclovers. In Nebraska, cooperative research with the Entomology Research Division traced most aphid-resistant, low-coumarin lines of biennial yellow sweetclover to germ plasm selected originally for large seed. These lines were also susceptible to leafhoppers and to blackstem disease and were early in maturity and low in forage productivity. A new pool of germ plasm was developed for an aphid-resistant, low-coumarin variety related to Goldtop. Selection of aphid resistance within low-coumarin annual and biennial white sweetclovers was continued. Plants highly resistant to simulated 2,4-D spray drift were selected within Goldtop and within Denta. Percentage of cross-pollination under field conditions for the variety Denta was determined using the low-coumarin gene, and was 55 percent. Cooperative studies established that cell-free extracts of sweetclover leaves in the presence of o-coumaric acid, uridine diphosphate, and B-mercaptoethanol catalyzed the formation of o-coumaric acid glucoside. Preparations from young leaves were much higher in glucosylating activity than those from older leaves; preparations of the cu/cu genotype were about 50% higher in activity than Cu/Cu leaf preparations. The B/b gene pair had no effect on glucosylating activity. Attempts to purify the glucosylating enzyme were unsuccessful. Distribution of B-glucosidase activity in centrifuged homogenates, between the supernatant fraction and the pellet, was influenced by plant environment, by pH and ionic strength of homogenate solution, and by adsorption of the enzyme on cellular debris. Ethylmethane sulfonate (EMS) at 0.003M and 0.004M concentrations was highly effective in producing chlorophyll-deficient and morphological mutants in a small, annual, autogamous strain of white sweetclover. Several EMS-induced variants appear to be single gene mutants. Callus cultures of various sweetclover lines were established.
- 2. Crimson Clover. Inheritance studies in Mississippi in F_1 , F_2 , and F_3 generations show the following characteristics conditioned by a single gene: (1) a male sterility caused by absence of anthers and completely associated with multiple ovaries and absence of petals, recessive to normal conditions;

- (2) glabrous leaves recessive to pubescent; (3) petiolulated leaflet recessive to sessile; and (4) multifoliolate leaf recessive to trifoliolate. Selections were made within S₃ lines for seed coat and plant and flower colors. Selection for non-shattering and high temperature dormancy of seed and for general agronomic performance of advanced inbred and polycross populations continues.
- 3. Alsike Clover. In Colorado, self-incompatibility of two clones of diploid alsike clover was changed to self-compatibility by the 4th day of 32° day-27°C night temperatures. Original compatibility was restored immediately at 21°C for one clone and within 24 hours for the second clone. The site of change in the latter clone was the style. The substance(s) responsible for the change in the compatibility reaction was not translocated from one flowering stem to another.
- 4. Red Clover. In Wisconsin, selection for persistence, for increased forage yields in second harvest year, and for resistance to powdery mildew, northern anthracnose, and viruses was effective. The Sf gene was incorporated into breeding materials to facilitate selfing. No new sterility allele was detected in the Syn2 derived from a population with 3 sterility alleles only; the Syn3 generation of this population was established in isolation. Studies measuring effectiveness of selection for large and small seed and estimating additive and nonadditive genetic variance in the variety Lakeland were initiated. A male sterile character appears to be simply inherited. Inheritance studies of several morphological characters were initiated; a dwarf character responded to application of gibberellic acid. World ecotypes did not vary greatly for pseudo-self-compatibility when evaluated at Beltsville, Md.; local ecotype Chesapeake exhibited highest PSC. Selection was continued for earliest and latest flowering red clovers.
- 5. White Clover. In South Carolina the interspecific hybrid T. nigrescens (2n=16) x T. occidentale (2n=16) produced F₃ plants indistinguishable from T. nigrescens. Embryogenesis of the F1 of this cross indicated slower development of fertilized ovules for first week, then much embryo abortion. Excising ovules at 7 or 14 days after pollination did not prevent abortion. Abortion appears due to hyperplasia of the endothelium of the inner integument. The cross T. nigrescens (2n=16) x T. occidentale (2n=32) was made with ease; the resulting triploids resembled T. occidentale and were highly sterile and non-stoloniferous. The cross T. repens x T. nigrescens was made at both diploid and tetraploid levels ($2n=32 \times 2n=16$ and $2n=64 \times 2n=32$); the resulting F1's varied in fertility, nodal rooting, and perennial habit. cross T. repens x T. occidentale was most readily made using repens as pistillate parent and tetraploid occidentale as pollen parent; most hybrids were perennial, stoloniferous, and exhibited normal chlorophyll. Many intraspecific crosses among accessions of T. nigrescens produced chlorotic F1 plants. Tolerance to root-knot nematodes, evaluated in the second year for 36 F₁ crosses of white clover, was attributable largely to general combining ability.

6. Berseem Clover. PL 480 research in Israel continued hybridizations between Berseem and closely related species. Most F_1 hybrid seed obtained failed to germinate or produced albino seedlings. Clover species were collected at 141 sites in eastern and western Turkey. Adaptation of clover species to soils of Israel was inconclusive; however, the study revealed a lack of potassium in some soils of the Upper Galilee. Additional diethylsulfate (DES) M_1 mutants of Berseem were obtained; M_2 lines were established from DES M_1 plants obtained last year.

B. Physiology and Biochemistry

- 1. Pollen tube growth enhanced in excised pistils of red clover. In Kentucky, di- or trisaccharides provided a better in vitro medium for pollen tube growth through excised pistils than did monosaccharides. Mannose inhibited pollen tube growth. Gibberellic acid, Naphthalene acetamide, and indole acetic acid at concentrations of 1, 10, and 100 ppm in the nutrient medium inhibited pollen tube growth in excised styles. Boric acid (10 to 100 ppm) and calcium nitrate (1,000 ppm) enhanced pollen tube growth; optimum temperature was 10 to 20°C. A prepollination effect of high temperature (40°C) in lessening degree of self-incompatibility was limited to styles and did not affect the pollen.
- 2. In vitro digestibilities of red clover clones determined. In Kentucky, 21 red clover clones varied from 52 to 59% digestible dry matter. These same clovers varied little for moisture percentage, indicating little association between DMD and moisture percentage of red clover clones. A modified DMD test was developed and does not require dichromate cleaning solution.
- 3. Axillary bud activity in white clover attributed to photosynthate translocation and accumulation. In South Carolina, white clover grown at 20°C produced branches at 73% of nodes; white clover grown at 30°C produced branches at only 20% of nodes. This plant material, differing greatly in axillary bud activity, was subjected to 14°CO2 for 1 hour and was then analyzed for general distribution of 14°C within plants via autoradiographs. Autoradiographs indicated that 14°C-labeled photosynthate moved rapidly from donor leaves and accumulated in sites of active growth, especially in terminal bud region and in young, expanding leaves. Definite activity was evident in axillary buds with 14°C-labeled photosynthate highest in plants subjected to 20°C.
- 4. Growth regulator effects in white clover seedlings. Indole acetic acid, gibberellic acid, and kinetin were applied singly and in combination with white clover seeds and seedlings in South Carolina to induce, possibly, qualitative differences in plant responses, especially a shift in the apical/axillary bud development pattern. Growth regulators at 10 and 1 ppm had striking effects on aseptically cultured seedlings: IAA greatly suppressed

root and leaf growth for first 4 weeks; GA produced elongated petioles and internodes with normal leaf and root development; K reduced leaf growth and number, suppressed root elongation, and increased axillary bud emergence and branch roots.

C. Quality and Variety Evaluation

1. Clovers. Two synthetics of red clover were superior in Wisconsin to Lakeland for persistence and for forage yields in second year of harvest. Kentucky Syn A-2 was especially persistent at the conclusion of the second harvest year in a Nebraska red clover variety test. Nebraska sweetclover tests indicate low-coumarin yellow sweetclover strains developed to date are earlier-flowering, and such strains produce about 1 ton per acre less dry matter than Goldtop. Goldtop sweetclover contributed 3 tons of dry matter and 189 pounds of nitrogen per acre in the fall of its first season of growth in Nebraska. Crimson clovers were superior to arrowleaf clovers in Mississippi for earlier growth and for total forage production as either an overseeded or volunteer legume in Coastal bermudagrass sod. Arrowleaf clovers were especially sensitive to spring defoliation.

-- Special Purpose Legumes --

RPA 208 - CONTROL OF DISEASES OF FIELD CROPS

- 1. Effect of virus on seed set in birdsfoot trefoil. A field study was completed in which three clones of birdsfoot trefoil infected with tobacco ringspot virus were compared with uninfected counterparts as related to characters affecting seed set. The virus-free plants had significantly greater numbers of (1) umbels per plant, (2) pods per umbel, and (3) seeds per pod, and differed in weight of 100 seed. A significant clone by virus interaction was shown for all characters except that of weight of 100 seed.
- 2. Factors affecting sporulation of Mycoleptodiscus spp. Studies on the factors affecting sporulation of Mycoleptodiscus terrestris and M. sphaericus were continued. These two fungi must have a natural medium on which to sporulate; i.e., intact plant cells. However, the cells need not be living. Fluorescent light regimes have been established and aeration requirements determined in order to produce conidia in volumes sufficient for large scale inoculations.

RPA 307 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY OF FIELD CROPS

A. Breeding and Genetics

1. <u>Trefoil</u>. Five trefoil varieties were evaluated in broadcast plots, drilled rows, and as spaced plants in the cooperative program at Blacksburg,

- Va. In this 2-year study, evaluation by any of the three methods gave the same results for the most important characters. F_1 hybrids from 56 inbreds have outyielded Viking by 50% in space plant trials at Ithaca. Improvement of trefoil through recurrent selection is continuing at Blacksburg, Va., Beltsville, Md., Columbia, Mo., and Ithaca, N. Y. A number of the recently selected synthetics continue to yield well. The effect of poor internal drainage on the productivity of trefoil and alfalfa was studied at Ithaca, N. Y. Four years average yields of Viking trefoil were 1.3 tons per acre greater than that of Narragansett alfalfa.
- Crownvetch. At Raleigh, N. C., 21 selected parental clones differing widely in morphology and maturity, and 100 crosses between these clones, are being evaluated. Average hay yields in 1967 revealed significant differences among individual crosses, among parental clones for progeny performance, and between different morphological types. In general, large-stem, large-leaf types were earlier and gave the highest yield the first cut. Small-stem, small-leaf clones yielded relatively better at subsequent harvests. crosses between large types and between small types were similar to yields of parental clones. However, the progenies of small x large and their reciprocal crosses were on the average equal to the large x large crosses, indicating some heterosis. At Beltsville, Md., clones and polycross progenies were evaluated under frequent close clipping to simulate continuous grazing. Erect, large-stem types were more productive than lower growing types in the early cuts: however, the latter were slightly more productive later in the season. After 2 years of frequent cutting, fair stands of many of the lowgrowing types remained while most of the erect-growing types failed to survive.
- Lupines. Studies conducted in the cooperative program at Tifton, Ga., in 1953-54 and again in 1967 indicated that early-maturing blue lupine varieties were 100% self-pollinated. These results and many other observations led breeders to expect 100% self pollination in the species. 1967 test designed to give maximum measurable cross pollination, the earlymaturing Rancher variety produced all self-pollinated seed (14,393). In the same test among 10,420 seed from a late-maturing line, 15 resulted from cross pollination. The results indicate the need of some isolation at least in the production of foundation seed of late-maturing varieties. A severe freeze in late February 1967 confirmed the superior winter-hardiness of 64-91 at Experiment (9°F) and Tifton, Ga. (17°F), and Gainesville, Fla. (18°F). Rancher variety winter-killed at all locations, whereas line 64-91 was undamaged. Five-hundred fifty pounds of breeder seed of 64-91 was produced in 1967. Release of 64-91 awaits the production of adequate foundation seed stocks.

In an informal cooperative breeding effort with the University of Western Australia, the resistance to seed shattering of the Australian variety 'Uniwhite' blue lupine was combined with anthracnose resistance, gray leafspot resistance, sweetness, soft seededness, and white flowers and seed of Rancher blue lupine. Seven F₃ progenies were found to be homozygous for all

of the desirable characters listed. This material should be a valuable basic breeding stock for Australia, South Africa, Europe, and the United States.

C. Quality and Variety Evaluation

- 1. <u>Silverleaf Desmodium</u>. <u>D. uncinatum</u> was established in association with Coastal bermudagrass in cooperative studies at Tifton, Ga. Yields the third year after establishment show that silverleaf has persisted well in association at 0- to 50-pound level of nitrogen, moderately well at the 100-pound rate, but was drastically reduced by the 200-pound rate of nitrogen. Better stands of silverleaf were maintained under a 3-week cutting frequency. Silverleaf significantly increased the yield of the mixture on plots receiving 0, 50, and 100 pounds of N per acre and may be useful as a summer growing perennial legume in association with Coastal bermudagrass on sandy soils in the lower South.
- 2. <u>Dolichos lablab</u>. <u>D. lablab</u> is one of the most productive summer annual legumes in the forage legume species screening tests at Tifton, Ga. Biotypes of this sub-tropic species tested prior to 1967 failed to set seed. Of 53 introduced <u>D. lablab</u> biotypes recently tested, 15 flowered and produced seed. The species is highly digestible and may well complement grazing programs with summer growing annual grasses.

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RPA 207 - CONTROL OF INSECT PESTS OF FIELD CROPS

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FORAGE GRASS AND TURF BREEDING AND GENETICS, DISEASES, QUALITY AND VARIETY EVALUATION, AND PHYSIOLOGY AND BIOCHEMISTRY

Crops Research Division, ARS

USDA and Cooperative Program

	:			Scienti	st	Man-Yea	ars	F.Y.	1968	
Location of Intramural Work	:-					h Prob			:	Total
	_:	207	:	208	:	307	:	906	-:	
	:		:		:		:		:	
Maryland (Beltsville)	•		•		:	.4	:	1.6	:	2.0
Arizona	:		:		:	1.0	:		:	1.0
Georgia		.6	:	.6	:	1.8	:	.5	:	3.5
Kentucky	:		:		:	1.0	:		:	1.0
Mississippi	:		:	.4	:	. 8	:		:	1.2
Nebraska	:		:		:	1.0	:		:	1.0
North Dakota	:		:		:	1.0	:		:	1.0
Pennsylvania	:		:		:	1.2	:		:	1.2
0klahoma	:		:		:	1.1	:		:	1.1
Texas	:		:		:	1.0	:		:	1.0
Utah	:		:		:	1.0	:			1.0
Wisconsin	:		:		:	1.0	•		•	1.0
Total	*	.6	:	1.0	:	12.3	:	2.1	:	16.0

Intramural program is supplemented by extramural support representing (a) 0.3 SMY's at State Agricultural Experiment Stations, 1/ and (b) P.L. 480 funds in 2 countries representing \$65,688 U.S. dollars equivalent.

^{1/} RPA 208

Problems and Objectives

Perennial grasses are grown on 1 billion acres for pasture and preserved feed. Yield improvement has been modest compared to that of annual crops. Breeding for improved forage quality and higher yield potential is technically feasible. Losses in production from pests on grasses go unrecognized or are accepted because alternatives for control are unsatisfactory. On turf alone, \$35 million is spent annually to control pests, reseed or resod where damages occur. Cost of turfgrass maintenance which exceeds \$4 billion annually, could be reduced 10% by development of improved varieties and maintenance practices.

Major objectives of the research are to develop and evaluate alternatives

- 1. Identifying and utilizing genetic variability in forage quality and yield.
- 2. Ascertaining reproductive characteristics of grasses and exploit their use in transfer of economically important traits.
- 3. Improving tolerance to physiological and environmental stresses such as drought, cold, heat, and harvesting.
- 4. Ascertaining and utilizing genetic basis for pest resistance.

Progress - USDA and Cooperative Program

-- Forage Grasses --

RPA 207 - CONTROL OF INSECT PESTS

Fall armyworm resistant pearl millet. Among 1,436 inbred pearl millet lines tested as seedlings at Tifton, Ga., 57 (4%) were rated resistant to first instar fall armyworm feeding. Larvae fed the resistant lines made slower growth than those which were fed the susceptible lines. In addition to having antibiotic properties, force-feeding on the resistant lines made larvae more cannibalistic.

RPA 208 - CONTROL OF DISEASES OF FIELD CROPS

1. <u>Hot water control of nematodes in bermudagrass</u>. Bermudagrass sprigs for shipping may be contaminated with endo- and ecto-parasitic nematodes. Research at Tifton, Ga., has shown that bermudagrass stems and stolons will partially tolerate immersion in water at 55°C for 45 minutes. Nematodes were killed by the treatment.

Hot-water-treated bermudagrass sprigs transplanted to a field nursery, newly treated with methyl bromide, can provide nematode-free planting stock if outside contamination is prevented.

2. <u>Diseases affect pearl millet</u>. Increase in leafspot and head mold injury is associated with advanced host age and greatly reduce seed yield of pearl

millet in the Southeastern United States. Susceptibility at the seedling stage is still a factor in forage production in the Southeast.

At Tifton, Ga., common pearl millet, Starr, a late experimental synthetic, Gahi-1, the 4 parent inbreds of Gahi-1, and the 6 diallel crosses were evaluated for resistance to Helminthosporium setaria. Disease reaction was most severe on 1-week old seedlings and least severe on 6-to 7-week old seedlings. The late synthetic experimental strain remained most resistant. In addition, Gahi-1 and 2 of the diallel crosses tended to remain resistant after the 6th week.

At Tifton seedlings escape seed rot, damping-off and leaf blight when planting is delayed until the onset of warm weather.

3. Alkaloids in tall fescue identified. At Lexington, Ky., research has shown perloline to be the major alkaloid in tall fescue. This alkaloid is known to be toxic to paramecia and certain warm-blooded animals. Forage sampled in August showed reduced level of perloline in disease-susceptible 'Ky. 31', 'Alta', and 'Goar' tall fescue when sampled in August. Very high level of perloline was found in 'Kenwell', a variety with high degree of tolerance to Helminthosporium.

RPA 307 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY OF FIELD CROPS

A. Breeding and Genetics

- 1. Chemically induced sterility of pearl millet. Research at Tifton, Ga., has shown that dimethylarsinic acid (DMA) as foliar spray at early growth stages causes gross morphological changes in floral structures of pearl millet. A significant increase of univalent chromosomes at meiosis was associated with a marked reduction in seed set following treatment.
- 2. Winterhardiness improved in buffelgrass. Progress in obtaining more winterhardy buffelgrass has been made at College Station, Tex. Among 2,500 segregating F_2 plants from sexual F_1 buffelgrass, 19 plants survived the winter at Knox City in north Texas. These and some winterhardy apomictic F_1 hybrids will be used in hybridization work to obtain productive, coldtolerant buffelgrass. If the present level of cold tolerance can be fixed by apomixis in high-yielding F_1 hybrids, a potential variety can be used on over 50 million acres in the low 15-inch annual rainfall area of Texas. Present buffelgrass varieties are restricted to 12 million acres owing to lack of winterhardiness.
- 3. Interspecific and intergeneric hybridization. Cytogenetic research at Logan, Utah, suggests that the current taxonomic treatment of Elymus canadensis and Sitanion hystrix are unrealistic in view of their close genomic and phytogenetic relationships to Agropyron. Lack of chromosome pairing in A. scribneri and E. junceus indicate that these species are unrelated.

At Madison, Wis., several morphologically unstable plants were observed among interspecific sorghum-sudangrass hybrids. Uniformity within divergent selfed progenies from the same morphologically unstable plant suggested selective advantage of parental type gametes. Similar behavior has been demonstrated in unstable polyploid bromegrass and timothy species.

- 4. Yield and quality improvement in johnsongrass. Studies at State College, Miss., to increase culm number, culm height and to obtain denser plant base have resulted in selections with 25% higher forage yield. Johnsongrass crossed to sorghum has given superior lines which produce 6 to 9 tons of dry matter per acre from two harvests.
- 5. Effects of inbreeding and self-fertility in polyploid grasses. At Lincoln, Neb., inbreeding depression was demonstrated in little bluestem and switchgrass. Plant vigor was restored in little bluestem by hybridization of inbred ecotypes. Likewise the two-line Syn 1 progenies of switchgrass yielded significantly higher forage than advanced-generation line-bred selections. Research at Mandan, N. D., showed that S₁ of self-fertile clones yielded 50% as much forage as the 2-clone cross between a self-sterile and self-fertile clone. Forage yield of the reciprocal cross was 25% lower than when the self-sterile parent was the seed parent. The difference indicated that the low-yielding cross contained a high proportion of selfed seed. On the other hand, research at Madison, Wis., has shown that experimental synthetics of smooth bromegrass and timothy in which parent clones were somewhat self-fertile, produced higher forage yield than experimentals from highly self-sterile clones. Polycross progenies from modestly self-fertile bromegrass clones were also more tolerant to leaf disease, and had better agronomic characteristics as compared to those from self-sterile clones of the same origin.
- 6. Reproductive systems in polyploid grasses. The apomictic breeding system offers potential for production of superior forage grasses with hybrid vigor. The application of genetic basis for this mode of reproduction has been most encouraging in breeding for improved forage production and winter hardiness of buffelgrass. Research at College Station, Tex., identified superior apomictic hybrids which have substantially outyielded the newly released Higgins and are more winterhardy.

Manipulation of apomixis is of utmost importance in several perennial forage grasses such as dallisgrass, bahiagrass, and buffelgrass.

7. Genetic stocks of pearl millet catalogued. A PL 480 project in India has provided the description of the presently available world collection of germ plasm. The plant material was evaluated in 13 different environments ranging from 2°N to 32°N latitude. African stocks were generally more pest resistant than Indian except for ergot resistance. Indian stocks were generally earlier in maturity and may have escaped infection.

B. Physiology and Culture

1. High carotene and xanthophyll from Coastal bermudagrass. In the Southeastern United States poultry rations have been supplemented with dehydrated alfalfa meal to supply adequate carotene and xanthophyll. Locally grown Coastal bermudagrass meal has been shown to be a satisfactory substitute for alfalfa meal. Research at Tifton, Ga., showed that excellent annual yields of 8.3 lbs/acre of carotene and 11.8 lbs/acre of xanthophyll can be obtained from Coastal bermudagrass fertilized with 750 lbs/acre of N and with P2O5 and K2O to make a 4-1-2 ratio. Forage cut at 24- to 25-day intervals averaged 17.5 to 18.0% protein. Fertilizer without P and K reduced the total forage yield by 45% without affecting protein, carotene and xanthophyll contents of the grass grown on Tifton loamy sand of medium-available P and low-available K.

-- Turfgrasses --

RPA 906 - CULTURE AND PROTECTION OF ORNAMENTALS AND TURF

- 1. Chemical control of cottony blight. In the South, cottony blight is the most severe disease on golf greens overseeded with annual ryegrass. Studies at Tifton, Ga., have shown that Terrazole (5-etoxy-3-(trichloromethy1)-1,2,4-thiodiozole at 8 oz. (35% WP) per 1,000 sq. ft. gave satisfactory control without causing phytotoxicity. Varieties of creeping bentgrass differed in reaction to the chemical.
- 2. <u>Kentucky bluegrasses vary in tolerance to sod webworm</u>. At Lexington, Ky., 'Kenblue' was more tolerant to sod webworm than other named varieties and common Kentucky bluegrass lots from Denmark and The Netherlands.

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-- Forage Grasses --

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RPA 307 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY OF FIELD CROPS

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RPA 906 - CULTURE AND PROTECTION OF ORNAMENTALS AND TURF

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PASTURE AND RANGE QUALITY, PHYSIOLOGY AND BIOCHEMISTRY, SEEDING AND ESTABLISHMENT, PROCESSING AND MANAGEMENT

Crops Research Division, ARS

USDA and Cooperative Program

	: Scient	ist Man-	Yea	rs F.Y. 1968		
Location of Intramural Wor	k : I	Research	Pro	blem Area	:	Total
	:	112	:	307	:	
	•		:			
Arid Pasture & Range	:		:		•	
Arizona	:	2.0	•			2.0
California	•	1.0	:			1.0
Colorado	•	3.0			:	3,0
Montana - Bozeman	•		:	1.0		1.0
- Miles City	•	1.0	:			1.0
Nevada	•	1.0	:			1.0
New Mexico	•	2.0	•		:	2.0
North Dakota		2.0	:		:	2.0
Oklahoma	:	1.8	•		:	1.8
Oregon	•	3.0	:		:	3.0
Utah	•	3.0	:			3.0
Washington	•	1.0	:		:	1.0
Total Arid Pastu	re &:Range	20.8	:	1.0	:	21.8
	•		:		:	
Humid Pasture & Range	:		:		:	
Maryland (Beltsville)	•		:	3.0	:	3.0
Florida			:	1.0	:	1.0
Georgia	•		:	1.0	:	1.0
Indiana	•		:	1.0	:	1.0
Minnesota	:		:	1.0	:	1.0
Missouri				1.0	:	1.0
North Carolina	•		:	1.0	:	1.0
Pennsylvania	:		:	1.8		1.8
Total Humid Past	ure & Range		:	10.8	:	10.8
Total	:	20.8	:	11.8	:	32.6

Intramural Program is supplemented by extramural support representing (a) 0 SMY's at State Agricultural Experiment Stations, and (b) P. L. 480 funds in 3 countries representing \$44,869 U. S. dollars equivalent.

Problems and Objectives

Forage and rangeland constitutes I billion acres of the agricultural land in the U.S. or three times as much as all other harvested crops. This land produces the majority of the feed for all ruminants as well as special feeds, such as dehydrated alfalfa for some nonruminants. Increasing world need for livestock products and smaller margin of profits to individual producer demands more intensive management. This can only be brought about by research directed toward improved establishment, specific management practices for new and improved varieties, and discovering basic physiologic and biochemical principles which can be used to develop improved varieties.

Objectives:

- 1. To increase probability of a successful seeding.
- 2. To maximize efficiency of solar energy conversion per acre of forage.
- 3. To increase the efficiency of harvesting, whether by animal or by machine.
- 4. To increase level and uniformity of forage quality.
- 5. To define principles of growth which can be used to develop improved varieties.

Progress - USDA and Cooperative Program

-- Arid Pasture and Range --

RPA 112 - RANGE MANAGEMENT

A. Quality

- 1. Light quality unchanged by saran screen shades. Saran screen was used to provide various degrees of shade on plots of <u>Bouteloua gracilis</u> at Mandan, N. D. A spectroradiometer was used to measure the energy rate intensity over the wave length range of 400 to 750 millimicrons to determine whether or not the saran screen altered spectral composition of the sunlight. Reduction in energy rate intensity was found to be uniform across the spectral range checked. There was little change in spectral composition as sunlight passed through the saran screen.
- 2. Paraquat-treated grass yields superior beef gains. Studies at Burns, Ore., have shown that standing crested wheatgrass can be cured by spraying with .2 lb paraquat/acre + 1/2% v/vX-77 in late June. Yearling heifers averaging 664 lb each gained .91 lb/head/day for a 75-day period beginning August 15 when stocked at a light rate on cured grass. Control lots, also under light grazing, gained .34 lb/head/day. Grass can be sprayed between May 15 and June 30 with little effect on yield. The principal benefit of paraquat treatment is in maintaining forage quality rather than in increasing yield of dry matter.

B. Physiology and Biochemistry

- 1. Drought recovery slow on arid Southwest rangeland. On the Jornada Experimental Range in southern New Mexico the 1951-57 drought was the most severe in recorded history. Nearly complete elimination of black grama occurred on deep soils. Recovery by 1967 has been only to about 1% of its predrought level, with all perennial grasses yielding 49 lb/acre. On shallow soils the 1951-57 drought reduced black grama to about 50% of its predrought level and perennial grasses in 1967 yielded 161 and 313 lb/acre in two separate pastures.
- 2. Crested wheatgrass seed activated at low water potential. A field planting in southeastern Washington of seeds treated with ³²P labeled Na₂HPO₄ was made in October. Seeds placed at 1 inch, synthesized traces of phosphate esters when the soil water potential was -60 to -70 atm. This synthesis was accelerated after a light storm when the soil water potential increased to -12 atm. Additional precipitation raising the potential to -1 atm stimulated further phosphate ester synthesis and 40% of the seeds germinated. Seeds placed on the soil surface synthesized phosphate esters during periods of precipitation and high humidity. These products were broken down during drought. Germination did not occur during the 32-day experimental period, but 1% of the seeds germinated during the relatively moist winter months. The water potential gradient of the surface inch of soil at one time was approximately 400 atm per inch. In such conditions the depth of seed placement can be highly important in the success or failure of range seedings.
- 3. Crested wheatgrass seeds synthesize ribonucleic acid at low water potentials. Four grams dry seed were coated with 100 microcuries of ³²P labeled NaH₂PO₄ and incubated at 23 C for 5 days over water or over saturated salt solutions having water potentials of -40, -130, or -370 atm in studies conducted at Pullman, Wash. Preliminary results indicate that these seeds synthesized ribonucleic acid at -130 atm but not at -370 atm. Microorganisms associated with seeds apparently did not contribute significantly to ³²P incorporation into ribonucleic acid at -130 atm. The objective of this research is to discover a physiological basis of drought tolerance.

C. Seeding and Establishment

1. Plowing most effective to seed pinyon-juniper range. Studies in north-central Arizona on land cleared of pinyon-juniper with an understory of blue grama have shown the importance of destroying all competing vegetation for success in range seeding. Plowing did this most effectively. In decreasing value were undercutting, strip undercutting, chemical fallow using dalapon prior to seeding, chemical fallow at the time of seeding and no soil preparation. Dalapon at 2 lb/acre applied in late May for early July plantings and in early July for mid-October plantings acted too slowly on blue grama sod to create an effective fallow.

- 2. Species widely adapted on pinyon-juniper range. Studies in north-central Arizona have shown western wheatgrass to be widely adapted and capable of withstanding considerable grazing pressure. Fourwing saltbush is the most promising browse species. Luna pubescent wheatgrass is promising but may offer management problems. Planting in furrows 2 inches deep has generally been superior to planting on flat surfaces.
- 3. Seeds of Atriplex polycarpa carry their own germination inhibitor. Germination studies conducted at Berkeley with seeds of Atriplex polycarpa, a valuable browse on arid lands in southern California, consistently gave slightly better germination from seeds leached with tap water than from seeds not leached. When leachate was used as the source of moisture, germination was drastically reduced. Germination was best at 15-20 C regardless of treatment.
- 4. Row spacing affects seed and forage yields differentially. Crested wheatgrass was seeded at Fort Collins, Colo, at row spacings of 6, 12, 18, and 24 inches and seeding rates of 10, 20, and 30 live seeds/foot of row. Stands obtained were approximately proportional to seeding rates. During the 5-7 years of the study highest yields were from 12- and 18-inch rows and lowest from 30-inch rows. The relationship between row spacing and forage yield did not change with time. Seed yield and seed stalk heights were directly proportional to row spacing. Seed yield was much more closely related to size of spikes rather than to numbers.
- 5. Control of wind erosion helps range seeding on Plains. At the Central Plains Experimental Range, Nunn, Colo., wind erosion has been stopped on sandy loam soils plowed for range seeding by packing the soil with a newly developed roller. The roller leaves ridges 4 inches high and 1 foot apart. It packs the plowed land to a bulk density comparable to preplowed conditions in the furrows (1.45 gm/cc) and to 1.25 gm/cc at the top of the ridges. Ridge erosion and furrow fill increased in proportion to the occurrence of high-intensity rain or hail but was unrelated to wind velocity.
- 6. Brush range in southern New Mexico yields to new conversion technique. Studies in southern New Mexico have shown that creosote bush range can be converted to grass by a combination of root plowing to destroy the brush and windrowing the destroyed plants onto the seeded grass rows to provide shade. The function of the shade is to reduce high soil temperatures, which grass seedlings cannot endure. Successful conversion is still highly dependent upon, and proportional to, summer precipitation and its distribution.
- 7. Grass seed treated before planting superior in field test. Seed of crested wheatgrass partially germinated and then dried was superior to seed not treated before planting in studies at Logan, Utah. In 13 field plantings made between May 9 and June 6 treated seeds gave a 13% better stand and when harvested yielded 14% more dry matter. In 6 plantings made between June 19

and July 20, stand from treated seed was 39% better than from untreated 28 to 39 days after planting. Seven days after planting nearly three times as many seedlings had emerged from treated as from untreated seeds. Environmental conditions during the two tests indicate that as conditions favorable to germination and seedling establishment deteriorate, the advantage of the preplanting seed treatment is enhanced. The data also suggest that in the field, in the arid west, conditions may never be ideal for seedling emergence.

8. Seeding successful on tarweed-infested ranges. Twenty methods of establishing grass on high elevation tarweed-infested ranges were studied in northern Utah and southern Idaho. Spraying in the spring with 2,4-D at 1-1/2 to 2 lb/acre and drilling in the fall with no additional seedbed preparation has proven successful and is used on large scale plantings in the National Forests. If the first spraying fails to control tarweed it is sprayed the following spring with 1 lb/acre 2,4-D.

D. Management

- 1. Water, first limiting resource, inefficiently used on rangeland. In the arid West, water is most generally the chief factor limiting plant growth. Studies at Nunn, Colo., reveal that in a favorable moisture year 75 lb forage, chiefly blue grama, will be produced per inch of growing season precipitation. This means that nearly a ton and a half of rain fell for each pound of dry matter produced. With the water requirement of blue grama at Nunn about 400 lb, it is evident that about 6 lb of growing-season rain are lost from the soil by runoff, evaporation, deep percolation or to undesirable vegetation to each pound contributing to the production of forage. In a more average rainfall year, with 89 lb forage per inch of growing-season rainfall, 5 lb are lost to each pound contributing to forage production.
- 2. Potential productivity of Central Plains ranges defined. With 400 lb of herbage per acre available to grazing cattle, studies at the Central Plains Experimental Range, Nunn, Colo., have shown the following mean values: 1 lb of beef for each 10.5 lb dry matter consumed; steers gain 2.2 lb daily and consume 23.2 lb dry matter; the forage has a net energy value 690 K cal/lb and a crude protein content of 12.7%. In May, when only 150 lb herbage is available, or in October, when moisture content of the herbage drops below 50%, conversion rates were less efficient. Growth of blue grama, the principal forage species ceases when moisture content falls below 41%.
- 3. Species respond differentially to season of grazing. Experimental pastures have been grazed 1 month each year at the Central Plains Experimental Range, Nunn, Colo., since 1963. Needle-and-thread (Stipa comata) has decreased as a result of close grazing during April, May, June, or July but has increased under close grazing during all other months. In contrast, blue grama, the principal forage species, has been unaffected by month of grazing.

- 4. Early season grazing of Russian wildrye may induce grass tetany. at Mandan, N. D., have revealed that Russian wildrye when grazed in spring and early summer may induce mild symptoms of grass tetany. Forage sampled daily from May 18 to June 19, 1966, had a ratio of K to (Ca + Mg) of 2.49 exceeding 2.2 in 27 of the 33 days, while in crested wheatgrass during the same period the ratio was 1.6 exceeding 2.2 on only 1 day. The Netherlands researchers consider forage having a ratio above 2.2 as tetany prone. 1966 steers on Russian wildrye gained 85 lb/head while those on crested wheatgrass gained 110 lb. In 1967 when the ratio of K to (Ca + Mg) was 1.9 for Russian wildrye and 1.6 for crested wheatgrass, respective gains were 89 and 99 lb/head. Accumulation of transaconitic acid in forage has also been linked with grass tetany. In 1966 between May 19 and June 18, Russian wildrye averaged 2.26% trans-aconitate while crested wheatgrass averaged 1.16%. Because of its nutritional value, wide adaptation, rapid recovery following rain, salt and drought tolerance, and long life, Russian wildrye is a very important species for western range improvement. Its possible tendency to be tetany-prone is, therefore, a serious problem.
- 5. Grass yields profoundly influenced by presence of sagebrush. Sagebrush which had reinvaded a crested wheatgrass range reduced grass yields in proportion to the abundance of sage. The study, begun in 1965 in southern Idaho, indicated in 1967 yields of 550, 850, 1042, and 1591 1b air dry grass per acre where sage (40" tall) was thinned to 20, 10, 5, and 0 plants per 100 ft². Whether sage was thinned by fire, chemicals, or grubbing was unimportant. Without sage removal winter moisture penetrated the soil no deeper than 24". With complete sage removal the soil was wet to 48". In a study at Burns, Ore, where sagebrush had reinvaded following control 15 years ago, no reduction in grass yield was evident where in rather dense stands of sage the sage was only 6" tall.

-- Humid Pasture and Range --

RPA 307 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY OF FIELD CROPS

A. Quality

- 1. <u>Digestibility as a selection criterion</u>. At Tifton, Ga., the "nylon-bag" and <u>in vitro</u> rumen fermentation techniques for measuring digestibility were compared for a wide range of small forage samples. The two methods correlated well except for low quality forages where the <u>in vitro</u> technique gave higher digestion coefficients. The "nylon-bag" digestibility technique was successfully used as a selection criterion in developing Coastcross-l bermudagrass. Either technique could be used by plant breeders to evaluate digestibility of forage plants.
- 2. Dwarf pearl millet plants are more digestible than tall plants. Near isogenic lines of tall and dwarf pearl millet were compared at Tifton, Ga., for in vitro digestible dry matter. Digestibility of leaves was the same for

both types but stems of the dwarf plants were more digestible. Steers fed dwarf millet forage ate 20% more and gained 50% faster than steers which were fed the tall millet forage. Although total yield of the dwarf type was less than the tall type, yield of digestible dry matter was similar for both.

3. Nitrogen needed for determining in vitro digestibility of high energy feeds. Laboratory studies in Minnesota show that urea added to in vitro rumen fermentation increases the digestibility of shelled corn, corn fodder, and sorghum fodder. Urea had no effect on digestibility of alfalfa. To samples which contain concentrates, this will permit direct comparison of digestibility of forage and concentrate ratios. Ten mg of urea per 250 mg of sample must be added for maximum digestibility.

B. Physiology and Biochemistry

- 1. Drought stress and the biochemical nature of reduced growth. At Gainesville, Fla., ribosomes of water-stressed plants functioned as well as those from control plants in the incorporation of amino acids into proteins. However, the soluble fraction from stressed plants was deficient in some essential component and incorporation was less in comparison to that of control plants. Water-stressed plants have higher levels of proline than check plants. When water-stress was released, proline uptake was higher than in the water-stress plant. Drought may slow growth by reducing synthesis of specific proline-rich proteins.
- 2. <u>Nitrate reductase activity in bermudagrass</u>. Coastal bermudagrass can use large quantities of nitrogen fertilizer, possibly because nitrogen metabolism is more efficient than in common bermudagrass. AtTifton, Ga., nitrate reductase activity of Coastal bermudagrass, receiving 800 lb/acre of nitrogen fertilizer, was considerably higher than from Coastal bermudagrass which received only 100 lb of nitrogen. Nitrate reductase activity of Kenya 56 bermudagrass was low and two Coastal by Kenya hybrids were intermediate.

C. Seeding and Establishment

- 1. Critical period defined for seedling establishment. At Beltsville, Md., yield of tall fescue the year following seeding could be predicted from the soil moisture available in the period from seeding to the first rain. During this critical period, decisions to reseed or irrigate could be made based on available soil moisture and number of dry days. Broadcast, or drill planting, produce comparable results when moisture was not limiting. When moisture was limiting, drilled plots yielded more.
- 2. Orchardgrass and birdsfoot trefoil suited to droughty soil. In southwest Missouri on upland droughty soils, a seeding of orchardgrass and birdsfoot trefoil mixture is more desirable than either by itself. The mixture has fewer weeds and higher total yields the first harvest year. In following years as the orchardgrass stand declined, birdsfoot trefoil continued to contribute to a high level of production.

D. Management

- 1. Self-fed steers on pastures efficiently convert energy supplement. Daily gains of steers on orchardgrass pastures at Beltsville, Md., can be predicted from forage available per steer. When molasses and urea was self-fed, gains increased about 80 grams for each kg of molasses. Energy supplement consumption per animal was the same regardless of the amount of forage available. At the highest stocking rate, efficiency of conversion of supplement was about equal to that achieved in dry lot feeding.
- 2. Reed canarygrass needs intensive management. In Minnesota, cattle gained 20% more per acre on reed canarygrass than on bromegrass pastures. Although reed canarygrass is generally thought to be less palatable, this is not a factor when it is properly managed. With intensive management, reed canarygrass returns greater profit per acre than bromegrass where grass-legume mixtures are not practical.
- 3. Pearl millet provides summer forage. In southwest Missouri, pearl millet is providing good quality forage during the summer months when cool-season perennials are unproductive.

Publications - USDA and Cooperative Program

-- Arid Pasture and Range --

RPA 112 - RANGE MANAGEMENT

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 - -- Humid Pasture and Range --

RPA 307 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY OF FIELD CROPS

Quality

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SEED CROP CULTURE AND MANAGEMENT, PHYSIOLOGY AND BIOCHEMISTRY, DISEASES, AND QUALITY AND VARTETY EVALUATION

Crops Research Division, ARS

USDA and Cooperative Program

	: Scientist Man-Years F. Y. 1968								
Location of Intramural Work	: Resear	: Total							
	: 307	208	304	6031/					
	•	•							
Maryland (Beltsville)	: 1.0	*	•	•	1.0				
Arizona	. 1.0	:	•	•	1.0				
California	. 1.2	:	•	•	1.2				
Colorado	•	:	1.8	•	1.8				
Indiana	1.0	:	•	•	1.0				
Oklahoma	. 1.0	:		•	1.0				
Oregon	. 1.0	: 1.0		•	2.0				
Utah	. 0.5	:		•	0.5				
Viet Nam	•	*		9.1	9.1				
Washington	2.0	•	•	•	2.0				
Total	: 8.7	: 1.0	: 1.8	: 9.1	20.6				

Intramural program is supplemented by extramural support representing (a) 0 SMY's at State Agricultural Experiment Stations, (b) 0 SMY's at other U. S. institutions, and (c) P. L. 480 funds in 4 countries representing 201,455 U. S. dollars equivalent.

^{1/} RPA 603 Plant and Seed Multiplication in Viet Nam. ARS/USAID PASA.

Problems and Objectives

Domestic production of grass and legume seed crops is approximately 750 million pounds, valued at \$175 million to the producers. To overcome deficits and to assure adequate supplies, 75 million pounds of seed are imported annually. This is necessary because technology has not been developed to make forage seed production a profitable farm enterprise. Practices used in the harvesting, processing, and storage of vegetable seeds often influences seedling emergence and uniformity of stand. Mechanization of the vegetable industry requires increase of speed and uniformity of seedling development. There is insufficient basic knowledge of physiology of seed development and germination as these influence the crop-producing potential of seeds and potential industry mechanization.

Major objectives of the research are to:

1. Elucidate physiologic phenomena and biochemical characteristics associated with floral development, pollination, and seed set in forages.

2. Develop efficient cultural and management practices to provide maximum reproduction potential in grasses and legumes, and develop improved methods for evaluating seed quality.

3. Formulate new methods for economically preventing or controlling diseases in grass-seed fields by altering cultural practices and evaluating new chemicals for fungicidal activity.

4. Identify the primary factors that contribute to population shifts in forages, and co-ordinate the production of foundation seed.

5. Discover mechanisms controlling germination and seedling vigor in vegetable seed.

6. Establish relationships between conditions of vegetable seed development and seed quality.

7. Utilize this information to produce vegetable seeds with higher and more uniform quality required for mechanized agriculture.

Progress - USDA and Cooperative Programs

RPA 307 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY

A. Culture and Hanagement

1. Seed buried in the soil remain viable for years. Results from cooperative research at Corvallis, Ore., show that seed of grass and legume species retain enough viability after 65 months to cause serious varietal contamination. The average live seed from 5 depths was 1.4% for annual ryegrass, 5.3% for crimson clover, 8.9% for Highland bentgrass, and

- 45.7% for red clover. Shortest lived in the soil were red fescue, perennial ryegrass, orchardgrass, and chewings fescue.
- 2. Higher axillary inflorescences produce the most seed in Eastern gamagrass. At Stillwater, Okla., studies with Eastern gamagrass have shown that the pistillate units of the terminal and first and second axillary inflorescences have the highest percent seed set. To obtain maximum seed yield and quality, harvest must be timed before disarticulation of the spiklets on the terminal and first and secondary axillary inflorescences.
- 3. Age of stand affects seed yield. At Pullman, Wash., seed was harvested from 1-, 2-, and 3-year-old plants of Kentucky bluegrass grown in 30-, 60-, and 90-cm spacings. The seed yield of the 2-year-old plants averaged 845 kg/ha whereas the 1- and 3-year-old plants averaged 548 and 578 kg/ha, respectively. The 30- and 60-cm spacings produced significantly higher yields than the 90-cm spacing. The highest seed yield for the 1-year-old plants was produced in the 30-cm spacing; the 2-year-old in 60-cm spacing; and the 3-year-old in 90-cm spacing. Quality of seed was influenced by age of plants. The 1-year-old plants produced the heavier seed and the 1- and 2-year-old plants had seed with higher germination than 3-year-old plants.
- 4. Seed losses in switchgrass. Cooperative studies at Stillwater, Okla., have established seed blasting largely responsible for low seed yields in switchgrass. Measurements on irrigated and non-irrigated stands of different age show that the percent of seed lost is uniformly distributed within a given year and is not exclusively related to age of plant.
- 5. Distribution of foundation seed of forage species. In 1967, 119,776 pounds of superior forage-crop varieties were distributed by the Foundation Seed Project in 15 States, Bolivia, Canada, and West Pakistan. Foundation seed totaling 121,210 pounds was produced in Arizona, Idaho, New York, Texas, and Washington. Verdant timothy and Tillman white clover were added to the Project. Foundation seed is being produced in Wisconsin, Minnesota, and Washington.

B. Physiology and Biochemistry

- 1. Volatile odor components of honeybee-collected pollen vary. In cooperative research at Tucson, Ariz., a survey of the volatile odor components of honeybee-collected pollens of 17 species was studied. The results from the gas chromatographic analyses show that the volatiles emanating from these pollens vary quantitatively, depending upon plant species, storage time and temperature, and moisture content.
- 2. Comparative plant growth under fluorescent and "Lucalux" lamp systems. At Lafayette, Ind., plant species were grown in the greenhouse under supplemental lighting provided by a fluorescent-filament system, and "Lucalux", the high efficiency, high pressure, multi-vapor lamp. Seedling growth of 17 species was as good or superior under the "Lucalux"

lamp. Two species, smartweed and pigweed, grew much better. The "Lucalux" lamp has potential as a supplemental light source in plant-growth chambers and in greenhouses.

C. Quality and Variety Evaluation

- 1. Small genetic shifts occur when seed of Finnish-bred varieties is multiplied in the U.S. Results from a P. L. 480 Project in Finland show that seed of Tammisto red clover can be produced, generally, for at least two generations without any detrimental changes in the variety. In third-generation red clover produced at Shafter, Calif., significant changes toward earlier types have occurred. In the Tepa tetraploid red clover genetic shifts have been smaller than in the diploid variety. However, changes have been small in the diploid Tammisto and tetraploid Iso alsike clover, even in the third generation. Second-generation Tammisto white clover produced in the U.S. has about the same winter hardiness as the original basic seed. Two and three generations of Finnish orchard-grass, meadow fescue, and timothy seed grown in the U.S., in general, show no detrimental changes. In the Shafter, Calif., grown meadow fescue seed, there has been an increase in the percent of early flowering plants.
- 2. Environmental pressures during seed increase result in changes in plant populations of birdsfoot trefoil. Cooperative studies at Lafayette, Indiana, of second and third generation birdsfoot trefoil seed produced at Shafter and Tehachapi, Calif.; Logan, Utah; Prosser, Wash.; Tikkurila, Finland; and Rehovot, Israel, have shown plant populations from seed lots produced at the four Western locations of the U.S. did not differ. Third generation seed produced in Finland and Israel showed population changes toward later flowering plants. Seed from Israel produced plants that were more susceptible to winter-injury. This loss of winter-hardiness was more pronounced in the third generation.

Similar studies were made with Empire seed produced in North Dakota, New York, Beaverlodge, Canada, and Israel. There were few differences in growth habit among the plant populations from seed from these locations. There was a trend toward earlier flowering among plants from the Beaverlodge increase. All first generation seed produced taller plants than did the original seed. The magnitude of the change was relatively small.

RPA 208 - CONTROL OF DISEASES

D. Diseases

1. Systemic fungicides provide disease control in grass-seed crops in lieu of burning. In studies at Corvallis, Oreg., a fungicide applied to the soil gave complete control of blind seed disease and ergot for the first time by a systemic fungicide. Flag and stripe smut diseases were eradicated or suppressed for long periods in Merion Kentucky bluegrass by chemicals with fungistatic properties.

RPA 304 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY

M. Physiology and Biochemistry

- 1. Snapbean germination. Germination of several newer snapbean varieties is extremely sensitive to stresses during the initial stages of germination. These stresses include low (15° f.) imbibition temperature, low (10%) seed moisture, and deficiency of oxygen. Older varieties are not sensitive to these stresses, thus these results account for the changing pattern of germination problems encountered in recent years.
- 2. Lima bean axis growth. In excised lima bean axes, the first 12 hours imbibition and growth is especially sensitive to lack of oxygen. This observation emphasizes previous data on the critical importance of the very first stages of germination. The enzyme ribonuclease develops parallel to growth of the embryonic axis, but appears not to control the initial temperature sensitivity.
- 3. Onion seed yield. Low seed yields in hybrid onions result from several biological processes, all associated with pollination: failure of male and female parents to bloom at the same time, failure of insect pollen transport, and a process involving either failure of pollen to fertilize the ovule or embryo abortion after fertilization. If the latter is actually embryo abortion, this indicates that more pollen is necessary for seed development than the single pollen grain required for fertilization. The fact that three types of failure produce the same visible symptoms accounts for previous confusion as to the mature of the problem.

RPA 603 - PLANT AND SEED MULTIPLICATION

Orop production in Viet Name. Under the AMS/USAID PASA over 200 varieties, hybrids, selections, and indigenous ecotypes of field and horticultural crops have been sent to Viet Nam. These are evaluated in various environments and management systems by the Agricultural Research Service, GVN, with assistance from U.S. Research Advisors. In addition to the U.S. seed was obtained from Mexico, Columbia, Guatemala, Philippines, Thailand, and India. Although an increase in rice production is the goal, the objectives include plants to increase feed crops for the production of more animal protein and vegetable crops for local consumption. U.S. research advisers were initially located at experiment stations throughout the country. Because of the lack of security, all advisers are assigned in Saigon to work with counterparts in the Agricultural Research Service, GVN, except for who remains near Phan Rang. Preliminary data from plant-introduction nurseries show a great potential for increasing crop yields. Plans to assist the Vietnamese with a seed production program were canceled due to a reduction in staff.

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WEED AND NEMATODE CONTROL AND EPIDEMIOLOGY

Crops Research Division, ARS

USDA and Cooperative Program

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	: Research Problem Area :							
Location of Intramuarl Work	:	102	:	208	:	209	-:	Total
Maryland (Beltsville)	:		:	0.4	:		:	0.4
Arizona		3.0	:		:	0.3	:	3.3
Arkansas	•				:	1.0	:	1.0
Georgia	:		:	0.9	:	0.2	:	1.1
Indiana	•		:		:	1.0	:	1.0
Iowa			:	0.5	:		:	0.5
Kansas	:					1.0	:	1.0
Minnesota	:				:	0.2	:	0.2
Mississippi	•				:	1.3	:	1.3
Missouri	:		:		:	1.0	•	1.0
Nebraska	:	1.0			:			1.0
Nevada	:	2.0			:		:	2.0
New York	:		:		:	1.0	:	1.0
North Carolina	:		:		:	1.0	:	1.0
0klahoma	•	1.0	:		:		:	1.0
Oregon	:		:		:	1.0	:	1.0
Texas	:	5.0	:	0.1	:		•	5.1
Utah	:	2.0	:	0.7	:		:	2.7
Washington	:	2.0	*		:	0.5	:	2.5
Total		16.0	:	2.6	:	9.5	•	28.1

Intramural program is supplemented by extramural support through use of P.L. 480 funds in 3 countries representing 62,638 U.S. dollars equivalent.

Problems and Objectives

Effective and safe weed control is essential to the continued production of sufficient food, feed, and fiber to meet the increased requirements of our rapidly growing population. All methods of control—cultural, mechanical, biological, and chemical—must be employed to minimize crop losses due to weeds. For the foreseeable future, however, herbicide control measures, alone or in combination with other methods, offer the greatest promise for economically and effectively controlling problem weeds. In addition to effectiveness, herbicide measures must be rigidly evaluated for their safety with respect to lack of contamination of harvested crops and environment, and absence of harmful persistence in soils at levels which affect succeeding crops.

Nematodes attack all crop plants, and cause varying damage and crop losses. Most plant-parasitic nematodes infect roots and other underground parts of plants, but some attack bulbs, stems, leaves, and flowers. Damage initiated by nematodes is often extended by bacteria, fungi, and viruses. Damage can be reduced by use of varieties resistant to nematodes, but only a relatively few are available.

Fundamental information on the basic nature of disease resistance in plants is urgently needed to aid in the development of disease and insect resistant varieties. For root diseases caused by fungal pathogens for which the use of fungicides may not be feasible, biological control mechanisms operative in the soil may be the most promising solution. Physiological and biochemical studies of soil micro-organism-plant pathogen relationship are required in order to uncover these control mechanisms and to permit their exploitation.

Major objectives of the research are to:

- 1. Discover new principles and develop new and improved chemical, cultural, biological, mechanical, and combination methods of controlling weeds in plantings of agronomic crops; also those on grazing lands.
- 2. Determine sites and mechanism of action of herbicides and the basis for their selective action in weed and crop species.
- Determine the effects of climate, cultural practices, and soil composition on herbicide performance, persistence, and selectivity.

- 4. Determine factors which influence the germination, growth, and development of major weeds and to evaluate various stages of growth for susceptibility to control measures.
- 5. Investigate the physiological relations of nematodes to plants, including nature of resistance.
- 6. Evaluate susceptibility and resistance of crop plants, and factors determining or modifying pathogenicity.
- 7. Determine interrelationship of nematodes and plant diseases.
- 8. Develop cultural and biological control practices, as related to crops, soil, and environmental factors, that will reduce nematode losses.
- 9. Improve chemical control practices and determine the toxicity of nematocides to nematodes and crop plants.

Progress - USDA and Cooperative Program

RPA 209 - WEEDS AND OTHER HAZARDS

A. Cereal Crops

1. Control of weeds in corn. Investigations in North Carolina indicate that the herbicide pyriclor will be useful for selective control of witchweed in corn. In addition, tobacco appeared tolerant to the herbicide.

In Mississippi, a combination of dalapon plus MSMA applied to the foliage of emerged johnsongrass before planting of corn controlled johnsongrass from both seedlings and rhizomes without injury to corn. Combinations of 2,4-D with either atrazine or linuron provided excellent control of grasses and broadleaf weeds when applied to emerged weeds before planting of corn.

In extramural research in Illinois, corn, soybeans, and wheat were grown continuously in several locations for 3 years. Herbicide-treated plots of corn and soybeans out-yielded those in which cultural practices only were used; wheat yields were not affected by method of weed control. Crop stands were not affected by herbicide residues in the soil within the 3-year period. Crabgrass and velvetleaf appeared to be increasing in proportion to other weeds in continuous corn where only atrazine was used. Morningglory increased over other weeds in soybeans where only amiben was used. Where herbicides were rotated, no weed species appeared to be increasing. Herbicides controlled the weeds more effectively than cultural practices alone.

2. Control of weeds in rice. Postemergence applications of molinate in Arkansas controlled grass weeds effectively and selectively in rice for the third successive year. This new herbicide was recommended for practical use for the first time in the spring of 1968. Mixtures of herbicides or combinations of different herbicides applied at different times continued to control weeds better than single herbicides applied once. An early application of propanil followed by 2,4,5-T at midseason controlled grass, broadleaf, and aquatic weeds more effectively than single herbicides which controlled either grass or aquatic weeds. Higher vields of rice were produced with the combination treatment than with the single treatments. Mixtures of propanil with either of two developmental herbicides (each chemical at half the normal rate) controlled grass as well as propanil at the full normal rate of application. KN3 controlled aquatic weeds effectively but mixtures of the KN3 with propanil provided better control than either herbicide used alone. Rice growing under stress from cool temperatures and low soil pH was injured by KN3 or mixtures of KN3 with propanil; rice grown in a favorable environment was not injured. Drill-seeded rice seemed more susceptible to injury from the KN2-propanil mixtures than water-seeded rice, especially on silt loam. Mixtures of propanil and 2,4,5-T at low rates, for the third consecutive year, controlled aquatic weeds in water-seeded rice as well as high rates of either herbicide applied alone.

B. Forage Crops Grown for Seed

1. Control of weeds in legume seed crops. Amine or ester formulations of 2,4-DB (0.5 to 1.5 lb/A) reduced the yield of seed from Louisiana S-l white clover when applied at any of three different stages of growth.

Paraquat and diquat controlled winter annual broadleaf and grass weeds in fields of perennial clover being grown for seed without injuring the clover.

In Washington, dodder seedlings were controlled effectively by vapors of CIPC or dichlobenil from the surface of the soil. This finding helps to explain why incorporation of CIPC does not increase its effectiveness for control of dodder.

2. Control of weeds in grass seed crops. Split applications of diuron in October and February controlled all of the annual bluegrass in Kentucky bluegrass seed fields without adversely affecting the seed-yield.

Seed yields of Kentucky bluegrass and orchardgrass were reduced significantly when crop residue was left on the field after harvest. Where the residue was removed mechanically, sufficient vegetative material remained to interfere with the activity of herbicides. Burning the plots after

harvest removed the interfering residue and made conditions favorable for control of weeds with herbicides. Burning of crop residue creates an air pollution problem. However, burning the residue in grass seed fields helps to control diseases and weeds.

One 1b/A of terbacil controlled downy brome and increased the seed yield of creeping red fescue and Kentucky bluegrass in central and eastern Oregon.

RPA 112 - RANGE MANAGEMENT

A. Weeds in Grazing Lands

- 1. Mechanism of selectivity of 2,4-DB between alfalfa and broadleaf weeds. The mechanism of alfalfa resistance to 2,4-DB results primarily from the synthesis of herbicidally inactive long chain chlorophenoxy compounds from the parent herbicide. Thus, production of 2,4-D in lethal quantities by beta-oxidation and subsequent translocation to sites of action is prevented. After the application of 2,4-DB to alfalfa in New York, (2,4-dichlorophenoxy) caproic and (2,4-dichlorophenoxy)decanoic acids were detected as well as limited amounts of (2,4-dichlorophenoxy)crotonic and (2,4-dichlorophenoxy) acetic acids. The methyl ester of (2,4-dichlorophenoxy)crotonic acid was rapidly reduced on the alfalfa leaf surface to the methyl ester of 2,4-DB and, subsequently, the aforementioned metabolites were formed.
- 2. Weed control in seedling and established legumes. Paraquat in combination with tillage operations before seeding of legumes in New York was determined to have considerable potential for controlling Agropyron repens and Cyperus esculentus as well as annual broadleaf weeds and grasses. Combination herbicide treatments of (a) EPTC + bromoxynil; (b) propachlor + 2,4-DB; (c) bromoxynil + dalapon; (d) 2,4-DB + dalapon; and (e) DNBP + dalapon, were outstanding in giving season-long weed control during establishment of legumes.

In Missouri, postemergence treatments of 1 and 2 1b/A of MSMA did not reduce yields of alfalfa and controlled lambsquarters (Chenopodium album), smartweeds (Polygonum spp.), foxtail, other warm-season grasses, and waterhemp (Acnida spp.), but failed to control ragweed (Ambrosia spp.). Yields of lespedeza forage were greater from plots treated with 1/2 and 3/4 1b/A of chloroxynil than yields of plots treated with 1/2 and 3/4 1b/A of bromoxynil or 1 1b/A of 2,4-DB. Yields and germination of lespedeza seed were not affected by up to 1 1b/A of chloroxynil, bromoxynil, 2,4-D amine, and 2,4-DB ester.

In an established stand of birdsfoot trefoil (Lotus corniculatus) in Missouri, 1 1b/A of atrazine, 1 and 2 1b/A of simazine, 1/2 and 1 1b/A of bromacil, and 1/2 1b/A of terbacil, applied in March, controlled yellow

rocket (Barbarea vulgaris), curly dock (Rumex crispus), weed bromegrasses, and Erigeron spp. without injury to the birdsfoot trefoil. Four 1b/A of CIPC and 2 1b/A of atrazine, applied in March, injured birdsfoot trefoil.

Coronilla varia established in 1966 in Missouri with 1 lb/A of trifluralin, 1 lb/A of benefin, and 4 lb/A of 64-296-B yielded more than 6,000 pounds of oven-dry forage in 1967.

3. Control of perennial herbaceous weeds. Seven studies, originally established from 1963-1967, in Nebraska on the control of western whorled milkweed (Asclepias subverticillata) were retreated with various herbicides in June 1967. Picloram at 1 to 4 lb/A adequately controlled western whorled milkweed in experiments established in 1964. However, plots treated in 1963 were becoming reinfested. Retreatments resulted in excellent control. Dicamba and 2,4-D were ineffective.

The application of picloram on western ironweed in Nebraska at 1/2 lb/A, or more, effectively controlled top growth soon after treatment and, subsequently, destroyed the roots and rhizome. Repeated annual treatments with 2,4-D are required for reduction in stands.

4. Control of downy brome in new seedings of perennial grasses. In Nevada the greatest reduction in density of downy brome (Bromus tectorum) in the seedbed resulted from spraying with paraquat followed by furrowing and seeding perennial grass. Least reduction in density of downy brome resulted from no weed control treatment and surface drilling. Seedling stands of intermediate wheatgrass (Agropyron spp.) varied from .3 plants per foot when surface drilled only to 3.4 plants when plots were sprayed with paraquat and furrow seedbed. Severe and rapid depletion of moisture occurred in the top 3" of soil during a short but critical period of growth when seedlings were small in the plots that were surface drilled only, while in the paraquat-furrow-drilled treatments, soil moisture was only slightly lowered from field capacity. Otherwise, soil moisture was adequate for plant growth at all depths until near the end of the spring growing season. Rising air and soil temperatures were related to increased growth of weeds and intermediate wheatgrass seedlings. Air temperature l" above the soil surface was not affected by treatments. Soil temperatures, especially daytime, were lower in furrows than under surface-drilled plots.

B. Brush Control

1. Mixtures and additives that increase herbicidal effectiveness. A range of ratios of picloram to 2,4,5-T (water-soluble formulations) demonstrated that the activity of 2,4,5-T was markedly enhanced when the overall herbicidal content consisted of 15% picloram. A greater proportion of this compound did not further increase activity on mesquite in Arizona. The herbicidal effectiveness of 2,4,5-T against greenhouse-grown mesquite

seedlings was not greatly altered by the addition of amitrole or MSMA to the water phase but was substantially increased with the addition of picloram.

A study of carriers for the picloram-2,4,5-T combination in Arizona demonstrated that a nontoxic oil emulsion (with a 2,4,5-T ester in the oil phase) was relatively ineffective. A 2,4,5-T amine combination with both herbicides in the water phase was more effective although the same herbicides in a straight aqueous carrier were even better. Most effective of all was the picloram-2,4,5-T combination in a carrier of 50% DMSO.

Experiments corroborated that significantly enhanced activity was obtained with phenoxy herbicides when carried in the aqueous solution of 40% or more DMSO. Diesel oil or nontoxic oil, combined with equal volumes of DMSO, proved to be effective carriers for a 2,4,5-T ester particularly when emulsified with a relatively lipophylic surfactant. Xylene and glycerine were less effective than the oils in this respect.

In Texas, recovery of 2,4,5-T from live oak (Quercus virginiana) tissue was increased when low levels of picloram were applied in combination with the 2,4,5-T. The ester of 2,4,5-T was recovered 30 days after treatment in root tissues of live oak plants sprayed with 2 lb/A of the 2-ethylhexyl ester of 2,4,5-T.

Honey mesquite seedlings and small greenhouse plants reacted anatomically in a similar manner to 2,4,5-T and to picloram. Phloem cell proliferation caused an increase in seedling collar diameter. In greenhouse plants, the stem tip curled and died. The hypocotyl and main root enlarged as a result of periderm and phloem cell proliferation. The new xylem cells, particularly the vessels, did not enlarge as much as normal cells.

2. Research related to absorption and translocation of herbicides. There was relatively uniform xylem translocation in juniper (Juniperus spp.) branches stored 24 or more hours in the laboratory before testing, indicating that seasonal variations in juniper translocation may not be due to leaf changes from aging but to factors which would affect stomatal closure.

The lower epidermis of shrub live oak (Q. <u>turbinella</u>) leaves have numerous tufted trichomes between veins and glandular structures on the veins. These may affect herbicide absorption by the leaf.

Mesquite seedlings were analyzed for the presence of sucrose, raffinose, and stachyose. Only sucrose was found. When ¹⁴C-urea was applied to leaves of mesquite seedlings, translocation from mature leaves was predominantly acropetally and from immature leaves predominantly basipetally.

- 3. Picloram in runoff water from treated areas. In Arizona, small amounts of picloram were detected in the runoff water collected in July (0.017 ppm) and August (0.009 ppm) 1967 from treatments made on chaparral at El Oso in May 1965. No picloram has been detected leaving the area treated in 1966 on the Bumble Bee. Forage from an area on El Oso which received 3.3 lb/A picloram contained 20-30 ppm picloram at the end of the initial growing season after treatment.
- 4. Control of junipers and oaks. Diesel oil added to picloram in a water carrier speeded initial responses of alligator juniper, Utah juniper, and shrub live oak. Responses of alligator and Utah junipers were similar to those produced by adding 2,4-D to picloram. Oneseed juniper responses were not affected.

Pelleted and granular formulations of picloram applied to the soil killed 95-100% of the top growth of gambel oak one year after application.

- 5. Macartney rose. Mowing Macartney rose (Rosa bracteata), in Texas, in July or October significantly reduced ground cover the following spring. When mowed in May, the ground cover of Macartney rose did not differ significantly from unmowed plants a year after mowing. Mowing in May or October did not change the amount of tops killed by picloram treatments but mowing in July reduced the amount of tops killed. Mowing in May significantly increased tops killed and decreased ground cover of Macartney rose when it was subsequently treated with 2,4-D.
- 6. Factors contributing to effectiveness of aerial sprays of herbicides. In central and eastern Oklahoma factors contributing to poor oak control with 2,4,5-T in order of importance were: (a) application date; (b) air temperature at spraying above 90 F.; (c) spray coverage of 16% or less as measured by sampling cards in the sprayed areas; (d) relative humidity 60% or less; (e) airplane swath width in excess of 50 feet; (f) precipitation of 2" or less one month before spraying; and (g) oak leaves in dry waxy condition.
- 7. Mesquite control. In Arizona, hand spraying field mesquite demonstrated that a 50-50 mixture of the triisopropylamine salts of 2,4,5-T and picloram is more effective than equivalent rates of either the butoxyethanol ester of triethylamine salt formulations of 2,4,5-T, and probably more effective than picloram alone.

Of several new herbicides evaluated as soil treatments for control of seedling mesquite, one proved to be particularly effective. It was the emulsifiable concentrate of 1,1-dimethy1-3-(3-[N-tert-buty1-carbamyloxy] = phenyl)urea. At rates ranging from 0.4 to 50 lb/A it caused greater overall herbicidal response than did fenuron.

RPA 208 - CONTROL OF DISEASES

A. Nematodes

1. Forage and range. The alfalfa stem nematode, Ditylenchus dipsaci, is an important pest of alfalfa in all Western States. It was especially important in 1967 due to the cold, wet spring. Estimated losses to hay in the State of Utah alone were \$1.5 million. Alfalfa selections from crosses made by breeders in Nevada and screened by nematologists in Utah contained selections that were simplex, duplex, triplex, and quadruplex for resistance to the root-knot nematode. Progress is being made in the isolation of quadruplex and triplex genotypes that have high-level resistance for the root-knot nematode, the stem nematode, bacterial wilt, spotted aphid, and pea aphid, along with winter hardiness. Recent research showed that resistance of alfalfa selections to the northern root-knot nematode, and to the alfalfa stem nematode, decreased with an increase in temperature from 20° to 30° C, with no difference in ability of the nematodes to penetrate resistant and susceptible alfalfa. This indicates that resistance in present varieties is temperature dependent. Also, relative humidity determines infection by the stem nematode because alfalfa crown buds were more heavily infected with stem nematodes at high humidities than at low. In field situations, infection was particularly high in Utah, where alfalfa was cut immediately after irrigation.

Cooperative studies in Alabama and Georgia indicate that much progress has been made in increasing and stabilizing root-knot resistance in Sericea lespedeza to 3 root-knot nematode species (Meloidogyne incognita, M. incognita acrita, and M. hapla). Seventy four Sericea lespedeza breeding lines from 24 families, were tested for resistance to these nematodes in Georgia. After 4 years of backcrossing, there are now 13 lines from five families possessing good resistance to the root-knot nematodes.

In a 6-year study, an organic nitrogen fertilizer source (activated sewage sludge) reduced the number of sting nematodes on Tifgreen Bermudagrass in Georgia more than did inorganic nitrogen (ammonium nitrate). Plant growth reflected the lower nematode infestations where organic nitrogen was used as a fertilizer source, thus explaining the long observed benefits of turf grass fertilization with organic sources of nitrogen. In other studies roots of turf grass, used for vegetative propagation, were freed of nematodes without damage to the grass by soaking cores of sod in hot water held at 50°C for 15 minutes.

2. Grain. Field tests with nematocides on the High Plains of Texas indicated that root-knot nematodes (Meloidogyne incognita acrita) reduced grain sorghum yields from 15 to 40 percent. The rate of root-knot nematode increased in grain sorghum fields is less than in continuous cotton, but damaging populations are maintained by growing all present

varieties of sorghum in rotation with cotton. Only slight resistance to root-knot nematodes was found in selections from 16 breeding lines of grain sorghum.

B. Epidemiology

- 1. <u>Small grains</u>. The lateness of arrival of the known aphid vectors of barley yellow dwarf disease was correlated with a low incidence of the disease. This correlation, as well as time of appearance of first disease symptoms, was accurately predicted in the regional forecasting service in Lowa.
- 2. Germination and survival of propagules of Thielaviopsis basicola in soil. Soil-amendments of alfalfa hay, corn stover, V-8 juice, chestnut tannin, and soybean lecithin stimulated germination of both endoconidia and chlamydospores. Maximum germination occurred 18 hrs. after amendment addition followed by lysis of the germ tubes. Alfalfa or corn amendments reduced the fungistasis of the soil to near zero then increased it to a level greater than that of nonamended soil in 2-7 days. Asparagine, casamino acids, glucose, NH4NO3, and zein did not overcome fungistasis, whereas yeast extract and soybean protein were partially effective. Spores germinated adjacent to germinating bean seeds in nonamended soil but did not germinate adjacent to germinating seed in soil amended with alfalfa or corn 7 days before planting.
- 3. Survival of soilborne plant pathogens in the Midwest. Studies made under contract at Lincoln, Nebraska, were aimed at determining the source of initial inoculum of Helminthosporium turcicum, the cause of Northern Leaf Blight of corn. Less than 1% of the conidia in natural soil stored outdoors and moistened to the normal moisture capacity each month were viable after 8 months. A high percentage of the viable spores in soil contained chlamydospores which germinated to initiate infection. Surviving conidia of H. turcicum in soil may be another important source of initial inoculum for this disease.
- 4. A PL 480 Project on a study of antagonists from soil and rhizosphere against phytopathogenic micro-organisms. Ophiobolus graminis, Helminthosporium sativum, and Fusarium spp. were identified as the most serious pathogens of wheat in Yugoslavia. Studies of the numbers of antagonists to these pathogens from the rhizosphere of healthy and diseased wheat plants were initiated.

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CORN, SORGHUM, AND SMALL GRAIN INSECTS Entomology Research Division, ARS

RPA 207 - CONTROL OF INSECT PESTS OF FIELD CROPS

USDA and Cooperative Program

Location of Intramural Work	Commodity	Scientist Man-Years FY 1968
Georgia	Corn	10.0
Iowa	Corn	6.5
Mississippi	Corn	3.0
Ohio	Corn	2.0
South Dakota	Corn	- 5.9
Iowa	New crops	1.0
Indiana	Small grain	3.0
Kansas	Small grain	1.0
Louisiana	Small grain	1.0
Michigan	Small grain	3.0
Montana	Small grain	1.0
North Dakota	Small grain	2.0
Oklahoma Oklahoma	Small grain an	d
	sorghum	2.0
South Dakota	Small grain	4.1
rotal Total		45.5

Intramural program is supplemented by extramural support representing (a) 1.2 SMYs at State Agricultural Experiment Stations, (b) PL 480 funds in one country totaling 7600 U. S. dollars equivalent.

Problems and Objectives

Many species of insects cause losses amounting to millions of dollars annually to corn, sorghum, and small grains. It is estimated that 25 species of insects cause an annual loss of \$900 million to corn alone. Some of the most destructive insects of grain are: European corn borer, corn earworm, greenbug, Hessian fly, wheat stem sawfly, cereal leaf beetle, and rice water weevil. Some progress has been made toward the solution of some of the insect problems encountered in the production of grain crops, but more effective, more economical, and safer insect control measures are needed. Reducing crop losses in grain due to insect damage could result in net benefits of at least \$1 billion annually.

Major objectives of the research are to develop and evaluate alternative ways to control insect pests through:

- 1. Research on their biology, ecology, and population dynamics.
- 2. Insecticides that will not leave residues harmful to animals, man, or beneficial insects.
- 3. Use of attractants, sterilants, or other new approaches,
- 4. Development of insect resistant varieties.
- 5. Methods to avoid or counteract insect resistance to insecticides.
- A. Basic Biology, Physiology, and Nutrition (11.5 SMY)

1. Corn

a. European Corn Borer. At Ankeny, Iowa, tests conducted with corn borer resistant inbred Oh 43, which was released by the Ohio Agricultural Experiment Station in 1949, indicated that this inbred line has not mutated and/or lost its resistance to corn borer. Lines of Oh 43 maintained at Iowa State University and Ohio Agricultural Experiment Station continued to show comparable resistance to the borer.

At Ankeny tests conducted from 1965 to 1967 to determine if biotypes of the corn borer are developing that can successfully attack resistant inbred lines of corn, indicated that no such biotype has developed on at least one highly resistant inbred, CI.31A.

Data collected in 1966 and 1967 showed that over 90% second brood larval mortality occurred on resistant inbred lines within three days after egg hatch. Larvae from parental stock reared continuously (over 30 generations) on a meridic diet (laboratory strain) were less capable of attacking susceptible inbred inline WF9 than larvae reared from a field-collected population (wild strain).

At Wooster, Ohio, a mass selection program, under diapause-inducing conditions, for a non-diapause strain of European corn borer has produced a strain which has an incidence of over 93% non-diapause.

The 1967 early spring survey indicated a population of 3,833 borers per acre. A late spring survey conducted after corn fields had been disked and seeded to oats showed an average of 1,050 borers per acre. The midsummer borer population in Boone County was 3,521 borers (third generation) per acre. The summer generation averaged 6,552 borers per acre. However, the post-harvest survey showed that early frost and harvesting operations had reduced the population 81%, to 1,250 borers per acre.

Tests conducted in bioenvironmental chambers with crosses between moths collected in Alabama, Maryland, and Minnesota showed that borers containing Minnesota genes under Montgomery, Ala., or Baltimore, Md., conditions will diapause. However, the same genetic compositions under Minnesota conditions will pupate. This indicates genetic environmental interactions are exerted in favor of the Minnesota genes under more southern conditions and in favor of the Alabama or Maryland genes under northern conditions. However, the progeny of the Minnesota male X Minnesota female cross failed to pupate to any degree at any of the three simulated locations. This agrees with field results reported in the past that the borer in Minnesota is a univoltine type. These tests also indicate that the day length and temperatures of south central Alabama may suppress pupation and cause diapause to occur.

Field experiments investigating the possible existence of biotypes of the European corn borer conducted at Ankeny, Iowa, Waseca, Minn., Portageville, Mo., and North Platte, Nebr., with borer populations from six geographical areas showed (1) a gradient of increasing survival from north to south for all six biotypes; (2) decreasing rate of pupation from north to south. (This is related to the decrease in day length.); (3) decreasing rate of development for all six biotypes from 55 days at Waseca (most northern location) to 38 days at Portageville (most southern).

- b. Southwestern Corn Borer. In 1967, the southwestern corn borer spread to four additional counties in Alabama, two in Tennessee, and six in Kentucky. No new county infestations were found in Mississippi.
- c. Corn Earworm. A study conducted during the summer of 1967 at Tifton to determine suitable crops for production of the corn earworm pupae showed that the production potential of several host plants was as follows: sweet corn, 19,600 pupae per acre; dent corn, 9,400 pupae per acre; cotton, 4,200 per acre; sesame, 2,800 per acre; and tobacco, 2,200 per acre.

Research conducted under contract by the University of Michigan indicated that light phasing will alter the circadians rhythms of insects, especially Heliothis zea.

d. Corn Rootworm. Laboratory studies conducted at Brookings, S. D., on oviposition preference of the western corn rootworm indicated: (1) soil sites composed of large particles were preferred over small particles;

(2) cracks in soil were preferred to large soil aggregates; (3) moist soil was preferred to dry soil; and (4) slumps of grass were preferred to either corn stalks or surface trash.

Studies of adult corn rootworm hemolymph proteins using polyacrylamide gel electrophoresis indicated a difference in major protein constituents between adult southern and western corn rootworms. Fatty acid analysis of Diabrotica eggs showed that the fatty acid composition of the lipids in all three species was qualitatively identical.

Thin-layer chromatographic analysis of lipids from southern, western, and northern corn rootworm eggs indicated that triglycerides were the predominant neutral lipid and the major phospholipids were phosphatidyl choline and phosphatidyl ethanolamine. Tests conducted at Brookings, S. Dik., indicated that maxillae inhibit feeding of the corn rootworm larvae in the absence of sucrose, a feeding stimulant.

Fifteen host plants, other than corn, have been found on which the western corn rootworm larvae can complete their immature stages and emerge as adults in the field. They are: sand lovegrass, weeping lovegrass, intermediate wheatgrass, barley, wheat, spelt, pubescent wheatgrass, tall wheatgrass, rice, green foxtail, yellow foxtail, foxtail millet, teosinte, eastern gamagrass, and Job's tears.

e. Miscellaneous Insects. At Tifton, Ga., investigations of the antennae sensors in the family Noctuidae and Saturniidae have demonstrated two groups of sharply tapered sensillae on the scape and pedicel of these moths. These sensillae occur in dome-shaped configurations and are distinguished by extremely thin walls and a heavy, doughnut-shaped mounting at the base. They respond to visible radiation. Recordings of these sensors, while irradiated with light, show a typical biphase nerve potential which feeds back into the antennae and can be detected at any point along the main stem of the moth's antennae.

Studies being conducted in Zaria, Nigeria, Africa, under the AID/ARS Major Cereals Project indicate that instead of one species of shoot fly, there are at least three different types, probably species, of shoot fly damaging sorghum and millet in Africa. Stem borers causing damage on millet, sorghum, and maize are Busseola fusca and Sesamia.

Studies being conducted under the AID/ARS Major Cereals Project in Uganda, Africa, have indicated that the most serious insect problem on maize is the maize stalk borer, Busseola fusca. Insects attacking sorghum are the shoot fly, Atherigona varia soccato, stalk borers, Busseola fusca, Sesamia calamistis, and Chilo zonellus, and the sorghum midge, Contarinia sorghicola.

2. Small Grain and Sorghum Insects

a. Rice water weevil. A technique has been developed for mass rearing the rice water weevil, Lissorhoptrus oryzophilus, in the greenhouse which will make it possible to conduct studies on the host plant resistance and insecticidal control during the winter months.

Field studies conducted in Louisiana on Trichogramma indicated that Trichogramma parasitized 99+% of eggs laid by Chilo plejadillus and Diatraea saccha: alis during July and August.

b. Greenbug. Fi 1d collected greenbugs from the Stillwater, Okla., area and areas in other States have been identified as biotype B. Biotype A, the original field strain, can no longer be found in the field.

Several differences in greenbug biotypes A, original field strain, and B, current field strain, were studied. In all cases biotype A fed in the vascular bundle, but biotype B fed in the parenchyma (mesophyll) area of the leaf. Insertion of the stylets in feeding was intercellular and in a curving path by biotype A, but it was intracellular and in a straight-line path by biotype B. Biotype B was capable of destroying wheats with resistance to biotype A. Differences in nutritional requirements of the two biotypes also were observed. When both were fed on the standard chemically defined diet, biotype A weighed an average .378 mg in 16 days, as compared no .472 mg in 16 days for biotype B.

c. Hessian Fly. A new Hessian fly biotype Race E was identified from a sample of Hessian fly collected from a Georgia population. It differs from the Great Plains race and Races A, B, C, and D, in that it is capable of infesting wheats having the H₃ gene for resistance and Turkey wheat, having no resistance, but it is incapable of infesting wheats having the H₆ gene for resistance and wheats such as Vermillion and Seneca, resistant to the Great Plains race but susceptible to Races A, B, C, and D. Race F, not yet identified in field populations, was developed in the laboratory by manipulating genes for virulence in the Hessian fly.

Race B was the only race present in a sample collected from an infested wheat field in east-central Missouri where Monon wheat has been growing for several years. In experimental plots in central Missouri, Race A was the predominant race with a small percentage of Race B present. In north-east Kansas the predominant race was Race A, whereas in central and western Kansas and the Great Plains race was predominant. Races A, B, and C were present in small numbers in western, northeastern, and central Kansas, respectively.

Samples from 450 certified wheat fields in 64 Indiana counties had average Hessian fly infestations of 6.1% for varieties Monon, Redcoat, Reed, Riley, and Riley 67, and 1.0% for Knox 62 and Benhur. All varieties were infested with Race B. During 1967 Hessian fly infestations in

Michigan averaged 19%. In Ohio the infestation averaged 2.2%. Only traces of fly were found in North Dakota. Race B was present in all States.

Tests conducted in South Dakota indicated that wheat yield in small field plots, artificially infested with larvae of Eleodes suturalis at planting time, can be reduced 10%.

Studies conducted on the effect of photoperiod on corn leaf aphid indicated that aphids lived longer and produced more progeny when reared on barley under long day condition compared with short day. On artificial diet, aphids produced more progeny under long day.

An outbreak of army cutworm occurred in South Dakota iuring 1968. A large proportion of the winter wheat and alfalfa fields in western South Dakota were infested with 1 to 15 larvae per square foot. Intensive spraying was needed to control the insect. A majority of the larvae pupated by May 20 and moths appeared in light traps installed to trace their movement by June 7. A high incidence of parasitism and/or disease was found in the immature forms.

d. Wheat Stem Sawfly. In the fall of 1967, estimates were obtained of wheat stem sawfly populations and damage in 20 counties of western North Dakota, using a system of sequential analysis of the sampling units. Percent fields infested ranged from 75 to 100%. Of 20 counties sampled, it was estimated that 16 counties had 100% of the fields infested. Greatest damage from sawfly stem tunneling and cutting occurred in the northwestern counties.

At Fargo, N. Dak., studies on the effect of wavelength of light on wheat stem sawfly oviposition and infestation showed that sawfly oviposition was associated with amount of light transmitted at wavelengths of 425 millimicrons or less. These wavelengths are included in the violet and ultraviolet portions of the solar spectrum. Increased transmission of solar radiation at wavelengths of 425 mm, or less, resulted in increased sawfly oviposition, infestation, and damage.

Evidence has been found that wheat plants reflect ultraviolet light.

Amount of ultraviolet light reflected by wheat plants appears to be a function of: wheat variety, wheat species, stage of development, and degree of stem elongation. Experiments are in progress to test the hypothesis that sawfly preference for oviposition sites is a response to amount of ultraviolet light reflected from the plant.

e. Cereal Leaf Beetle. Recent research at East Lansing, Mich., has shown that diapause of the cereal leaf beetle can be terminated by topical treatment with the synthetic juvenile gonadotropic hormone (trans trans 10, 11-epoxyfarnesenic acid methyl ester). This is considered to be a major breakthrough in the rearing of the cereal leaf beetle as previous techniques required that adult beetles be held for about 90 days at 38° F to break

diapause. Laboratory studies indicate the possibility that only the female cereal leaf beetle has an obligatory diapause.

Laboratory studies conducted at East Lansing, Mich., revealed that light had a marked effect on oviposition behavior of the cereal leaf beetle. Oviposition is obtained after as little as four weeks of cold storage when beetles are kept in a 16-hour photoperiod; whereas, as much as 16 weeks of storage is needed before oviposition occurs under the other photoperiodic conditions.

Studies have indicated that the egg stage of the cereal leaf beetle is the most resistant stage to cold temperatures, followed by the pupa, larvae, and active summer adult, respectively. Complete development from egg to adult requires an average of 92 days at 58° F, 52 days at 67°, and 28 days at 80°, 23 days at 90°. An age specific life table constructed from laboratory populations of the cereal leaf beetle showed that mortality was highest in the pupa stage (39%), followed by the egg (35%), and the larval stage (33%). The net reproduction rate was calculated to be 3.9 under the laboratory rearing conditions.

Studies indicate that the cereal leaf beetle can be effectively sterilized by beta gamma and X-ray irradiation. 2,000 r appears to be the minimum amount of radiation required to sterilize the adult beetle. Histological changes in the neurosecretory system of the cereal leaf beetle indicated that diapause in Oulema melanopus is a result of an inactive corpus allatum.

f. Miscellaneous Insects. The lesser cornstalk borer was reared successfully for the first time on a wheat germ artificial diet. Egg, larval, pupal, and adult development were adequate for mass production procedures. The sorghum webworm was likewise mass reared for the first time on a similar wheat germ diet in the laboratory. This diet was also successfully used to rear all forms of the sugarcane borer.

The long life cycle of most spiders studied may be a limiting factor in their mass production for use in biological control of sorghum insects. Phidippus audax, for instance, required about 300 days to complete development from egg to adult.

Fall armyworms reared on the artificial wheat germ diet for 17 generations fed readily when transferred to seedling sorghum plants, and damage to the plants was comparable to that caused by larvae reared on natural food (sorghum).

- B. Insecticidal and Cultural Control (5.1 SMY)
- 1. Corn Insects
- a. European Corn Borer. At Ankeny, Iowa, systemic crop protection chemicals, American Cyanamid CL-47470 (4 pounds per acre) and carbofuran

(4 pounds per acre) applied at planting time produced good corn leaf aphid control 70 days after treatment. These chemicals plus TD-5032 exhibited encouraging borer control. Fourteen compounds tested against first generation borers at various rates, mixtures, and in various formulations indicated that EPN, parathion + heptachlor, parathion, diazinon, Azodrin, Gardona, Velsicol VCS-506, Hercules 14503, Union Carbide UC-30045, and DDT are effective insecticides for corn borer control.

At Ankeny, Iowa, the second year of examining the effect of visible phytotoxicity from the application of diazinon on yield and quality of 20 inbred and 45 single cross corn varieties substantiated previous results. Differential phytotoxicity was obtained with no reduction in yield, test weight, or kernel weight. Genetic and environmental factors influenced foliar expression of phytotoxicity to diazinon.

Experiments conducted to determine the effect of plant population (13, 17, and 26 thousand per acre) and row width (15, 20, 30, and 40 inches) on borer control, and borer establishment showed differences in control or establishment for first- or second-generation borers.

Laboratory studies designed to select for borer tolerance to a chlorinated hydrocarbon (DDT), organophosphate (diazinon), and carbamate (carbaryl) crop protection chemicals indicated that after 12 generations of selection pressure the borers developed a degree of tolerance to each chemical tested. A slight cross tolerance between classes of chemicals was also noticed.

The toxicities of phorate and five of its metabolites to the borer were determined to provide the relationship between residues present and insect control. Residues of these compounds were determined for the various parts of the plant treated with 1 pound actual per acre 1, 3, 7, 14, 21, and 28 days after application and at harvest. Phorate, its sulfoxide, and sulfone were the most toxic to the borer and were present in the whorl, as determined by chemical analysis, in the greatest amounts.

b. Rootworms. At Brookings, S. Dak, a 3-year study showed that removing corn for silage by September 1 resulted in significantly higher yields the following year due to a reduction in rootworm populations and corn lodging. There was an increase of 12.2, 11.0, and 11.6 bushels per acre for 1965, 1966, and 1967 following early silage removal. This indicates that heaviest oviposition occurs after September 1 in South Dakota, and that the use of chemical control might not be necessary the following year in fields where silage has been cut by this date.

The effect of environmental factors on the overwintering eggs of northern and western corn rootworms was evaluated in the field using spring and fall plowing as a means of relocating the eggs vertically in the soil. Winter temperatures below -8° C were lethal to corn rootworm eggs. Dessication of eggs near the soil surface due to lack of late fall and winter precipitation resulted in a greater egg mortality than that resulting from the cold temperatures.

c. Miscellaneous Insects. Research conducted under contract by the National Cash Register Company, produced several microencapsulation formulations of foliar and soil insecticides (malathion and diazinon) and viruses (specifically, polyhedrosis virus) for testing against alfalfa weevil, corn borer, grasshoppers, corn earworm, cotton bollworm, and corn rootworm. Preliminary results indicate little or no increase in control using encapsulated materials.

2. Small Grain Insects

a. Greenbug. At Stillwater, Okla., spray applications of Dursban to small grains for control of greenbugs at rates of .25 and .125 pound per acre resulted in 96 and 99% control after seven days. Of 24 other new insecticides tested for greenbug control, Dasanit, Bay 78182, Geigy GS-13005, Bomyl, and Thiocron were as effective as parathion at comparable application rates.

Liquid disulfoton used as a seed treatment for greenbug control on wheat, oats, and barley was very effective for 13 and 8 weeks for the .25 and .125 pound per 100 pounds seed treatment rates. Seed germination was reduced at higher rates of application. This material shows promise as an insecticide for greenbug control. Liquid disulfoton at varying rates in combination spray shows good promise for spring greenbug control in small grains.

- b. Cereal Leaf Beetle. At East Lansing, Mich., Bay 39007 and carbofuran gave excellent field control of adult cereal leaf beetle and larvae both as a seed treatment and as granular applications. All seed treatment rates of Bay 39007 (1, 2, and 4 ownces per 100 pounds of seed) and carbofuran (4 and 8 ownces per 100 pounds of seed) gave good control of spring adults and larvae. Granular application of one half pound per acre of Bay 39007 and carbofuran were also effective. Similar rates with Union Carbide UC-21149 killed significantly fewer larvae but the one pound rate was not different from Bay 39007 or carbofuran.
- c. Rice Water Weevil. At Baton Rouge, La., 20 insecticides were evaluated for control of the rice water weevil as seed treatments. Dursban was the only material that gave satisfactory control and did not show observable signs of phytotoxicity.

Carbofuran as a granular formulation applied at 0.5 pound active ingredient per acre 24 hours pre- to five days post-flood provided excellent control of the rice water weevil. Chevron RE 5305 at one pound active ingredient per acre also provided satisfactory control.

Studies on the effect of time of application on control of the rice water weevil indicate that the application of granular insecticides on the soil surface just hours prior to flooding gave the best maximum and long lasting control. Treatment with 0.5 pound active ingredient of carbofuran was very effective against weevil larvae established in the root systems of the seedlings when applied within 16 days after flooding.

C. Biological Control (4.1 SMY)

1. Corn Insects

a. European Corn Borer. At Ankeny, Iowa, Bacillus thuringiensis formulations 90TS(S7-145), liquid, S7-144; liquid with a protective agent, and S7-147 (microencapsulated preparation), were tested in the laboratory and field as microbial insecticides for control of the European corn borer. Applications were made on a basis of viable spores per gram of formulation. At 25 x 10⁶ viable spores per gram of diet, S7-145 and S7-146 formulations gave 79.5% and 89.0% mortality after 48 hours. S7-144 gave 41.1% mortality.

In field studies S7-144 gave 76% control; S7-145, 51%; S7-146, 61%; and DDT (check), 72%.

Horogenes punctorius and Macroncentrus grandii were the two prominent parasite species collected in a survey covering nine States in the Midwest. A few Pyraustomyia penitalis and Lydella thompsoni were also collected. The highest parasitism was 12.8% by M. grandii in North Dakota. No Trichogramma sp. were detected.

Perezia pyraustae infection rates in Boone County, Iowa, during the 1967 surveys were the lowest ever recorded. During this same period there was a slight increase in corm borer larvae per acre over the previous year.

At Brookings, S. Dak., seven chlorinated hydrocarbons and organophosphates were tested in vitro for compatability with B. thuringiensis. Growth curves were established for nutrient broth cultures containing 0.7, 1.7, and 3.5 ppm (equivalent to 1, 2.5, and 5 pounds per acre actual) insecticide. Bacterial growth was prohibited with 3.5 ppm DDT, aldrin, and heptachlor. 1.7 ppm of the chlorinated hydrocarbons reduced bacterial populations by more than 50%. Growth curves were affected to a lesser degree at the 0.7 ppm level. Malathion, diazinon, and the epoxides of aldrin and heptachlor did not appear to affect growth when compared with the controls.

b. Fall Armyworm. At Lafayette, Ind., a histopathological study of two virus diseases of the fall armyworm, Spodoptera frugiperda, revealed that the granulosis virus attacks the fat body, causes a proliferation of cells, and requires a relatively long time to produce mortality; however, the nuclear polyhedrosis virus attacks a wide variety of tissues, does not cause a proliferation of cells, and produces mortality in a relatively short period of time.

A study was conducted at Tifton, Ga., to determine the effect of age and fertility of host eggs on their suitabllity for parasitism by T. evanescens. The study demonstrated there is very little or no variation in the suitability of Cadra cautella eggs during the first 60 hours of the incubation period. All the host eggs in this age range are equally suitable for oviposition and development by the parasite. During the last 12 to 24 hours

of the incubation period and associated with the period during which the morphological characteristics of the head of the host become evident, the eggs become progressively less acceptable for oviposition and development by the parasite.

Eggs from virgin females were often found to be suitable hosts for the parasite. However, the percentage of these eggs that were suitable varied considerably. The causes for the variation were unclear.

Eggs of one to five days of age from tepa-sterilized fall armyworm moths were found to be equally susceptible to parasitism by T. evanescens as one-and two-day-old control eggs. Since normal fall armyworm eggs usually hatch on the third day, chemosterilized eggs are available for parasitism at least twice as long as the normal eggs.

c. Corn Earworm. In feeding tests at Stillwater, Oklas, involving adults of ladybird beetles, lacewings, (beneficial insects), corn earworms, and 5th and 6th instar corn earworm larvae (destructive insects), the jumping spider, Phidippus audax, showed preference for the corn earworm larvae over a 7-day feeding period. Some of the beneficial insects were eaten. Egg laying female spiders were the heaviest feeders.

2. Small Grain and Sorghum Insects

- a. Army Cutworm. At Brookings, S. Dak., a rather large (120 x 240 m μ), single, rod-type virus embedded in polyhedral bodies measuring 2.5 μ in diameter was isolated from field collected larvae of the army cutworm. Preliminary studies indicate that only the fat body cells were infected with the virus.
- b. Cereal Leaf Beetle. During the spring and early summers of 1966 and 1967 field releases of Anaphes were made in various areas in southern Michigan. Several recoveries (parasites) were found in the spring of 1968, several miles from the release areas, indicating that Anaphes is now established in the cereal leaf beetle area.

A microbial analysis on several dying cereal leaf beetle larval specimens observed at East Lansing, Mich., revealed two potential pathogenic bacteria, Streptococcus faecalis var. liquefaciens, and Serratia marcescens. These as well as Aerobacter aerogenes and Proteus sp. which were previously reported from the cereal leaf beetle and later isolated from our laboratory cultures are also potential pathogens. The presence of these microorganisms suggest a strong possibility that the primary cause of mortality is linked with suboptimal environmental or rearing conditions.

Investigations conducted in Poland under PL 480 have led to the discovery of five additional parasites of Oulema gallaeciana overall parasitism recorded varied from 10 to 50%. A convenient morphological character of sexing cereal leaf beetle adults was found.

Several shipments of parasites collected under PL 480 in Yugoslavia were shipped to the United States for release in the cereal leaf beetle area. These shipments included about 4000 egg parasites (A. flavipes) and 3600 larvae parasites (unidentified).

- D. Insect Sterility, Attractants, and Other New Approaches (2.9 SMY)
- 1. Corn Insects
- a. Fall Armyworm. At Tifton, Ga., research was continued on sex pheromones of the fall armyworm, S. frugiperda. The active principle of the female fall armyworm was isolated in pure form and identified. When the synthetic was released in the greenhouse at the rate of one microgram per square yard for 10 days, it confused the adults to such a degree that mating and larvae damage was significantly reduced.

At Tifton, Ga., mating studies in which sterilized and fertile fall armyworm males were mated to normal females showed that the sequence in which the matings occur is very important. Females mated to a sterile male and then to a fertile male, oviposited 95% viable eggs, indicating that sterile sperm are not competitive with viable sperm. However, in the opposite mating sequence, results were not so clear. Oviposition from females mated to fertile males and then sterile males indicate an all or nothing type response. Approximately 45% of the females produced all sterile eggs after the second (sterile) mating while 45% produced all viable eggs.

- b. Corn Earworm. One hundred percent sterility was obtained on laboratory-reared corn earworm pupae one to two days from eclosion, with irradiation dosages of 17.5 KR (females) and 33.0 KR (males). Adults, less than one day of age when irradiated, were also sterilized with dosages of 17.5 KR (females) and 33.0 KR (males). At Tifton, Ga., Cobalt-60 was used to sterilize male and female fall armyworms. Adults of both sexes were effectively sterilized with 20 KR. Competitive trials show copulation uneffected by these treatments but sperms from treated males are not competitive with sperms of untreated males.
- c. Southwestern Corn Borer. At State College, Miss., studies on the effect of different levels of gamma radiation on the emergence of the southwestern corn borer from the pupa indicated that the early pupal stage is much more sensitive to radiation than either the mid- or late-pupal stages. The early pupal stage, one to two days old, cannot tolerate more than 5 KR without serious damage. Mid- (4 to 5 days old) and late- (7 to 8 days old) pupae can withstand levels as high as 20 KR without serious injury.
- d. European Corm Borer. A sex pheromone of the European corn borer has been isolated from 150,000 female moths. The male moth is apparently extremely resistant to the sex pheromone.

2. Small Grain Insects

- a. Cereal Leaf Beetle. Studies conducted on the cereal leaf beetle at East Lansing, Mich., indicated that mating was not seriously diminished by exposure of adults to 2000 r. Treatment levels of 4000 r and above of beta and X-rays reduced mating to a very low level. Egg hatch is affected by exposures of 500 and 1000 r, but no drastic reduction occurs until males are exposed to 2000 r. Survival was reduced by a factor of approximately one-half at 2000 r of beta and gamma radiation, but for the X-ray treatments the average survival time in days at 2000 r exposure level actually exceeded that in the controls. Reduction of 72.1, 72.5, and 84.4% were obtained in over-all hatching at the 9:1 ratio (male sterile to untreated females) for beta, gamma, and X-ray treated beetles, respectively. These tests would appear to indicate that, provided mass rearing can be achieved, the sterile male technique can be used to suppress or eradicate light cereal leaf beetle populations.
- b. Southern Corn Rootworm. Fewer eggs aere laid per female and egg viability was decreased when southern corn rootworm beetles were treated with apholate at 50 or 100 ppm either feeding or contact. Decreased egg viability is believed due to failure to transmit sperm to the female during copulation because of lack of seminal fluid.

E. Evaluation of Equipment for Insect Detection and Control (0.5 SMY)

A portable, battery operated, self-contained, recording weather station has been constructed for field use at Brookings, S. Dak. Ten sensor inputs record automatically for five minutes of every hour. Sensor inputs are for light intensity, wind speed, wind direction, rainfall, temperature (4 sensors), and humidity (2 sensors). Solar cells keep batteries charged. A roll of chart paper will contain the hourly recordings for 14 days.

Modified Steiner traps containing virgin females and STIKEM were used to survey natural populations of corn earworms and fall armyworms on the Island of St. Croix. A height of three feet was determined to capture significantly more numbers of males than any other height. Virgin females three days old were most attractive. Fed females caught significantly more males than non-fed females, and virgin females caught significantly more males than mated moths.

F. Varietal Evaluation for Insect Resistance (17.7 SMY)

1. Corn Insects

a. European Corn Borer. Tests conducted at Ankeny, Iowa, have shown that DIMBOA is a major factor in the natural resistance of corn to the European corn borer. A correlation coefficient of 0.76 has been established in plants between the concentrations of DIMBOA and field ratings for corn borer resistance levels. This indicates that a chemical analysis of plant

tissue for DIMBOA can be used to accurately predict the level of corn borer resistance in inbred and single cross hybrids.

Analytical results of seed at germination show that high concentration of two benzoxazinones, DIMBOA and a second benzoxazinone (DIBOA) occur in the embryonic corn plant. The endosperm and scutellum contain lower concentrations of the compounds. Biosynthesis of the benzoxazinones takes place throughout development of the plant; however, overall whole plant concentrations decrease as the plant matures. DIMBOA and DIBOA occur in all plant portions. Concentrations were generally highest in the roots of the plants, followed in decreasing order of concentration by the stalk, whorl, and leaf.

Greenhouse tests conducted with first instar larvae placed on first brood resistant and susceptible inbred corn plants show that larval migration is greater from resistant plants than from susceptible plants. Susceptible plants sprayed with the compound DIMBOA were also more repellent to the borer than unsprayed plants. These tests indicate that DIMBOA may be a non-preference chemical factor.

At Wooster, Ohio, 18 of 694 corn lines, from Michigan State Agricultural Experiment Station, indicated a good degree of resistance to European corn borer and leaf blight. Eight of 35 inbred lines tolerant to maize dwarf mosaic had a satisfactory degree of corn borer resistance. Nineteen of 59 S3 lines from the Ohio breeding material had a good degree of resistance.

In continuing tests at Ankeny, Iowa, to locate additional sources of first-brood resistance, two of 33 South Carolina inbred lines, and one of 14 Georgia inbreds had a good degree of resistance; 17 of 300 plant introductions had a good degree of resistance.

Several lines from a WF9 synthetic, B14 snythetic, 2-ear synthetic, corn borer synthetic #3, and other sources had a good degree of resistance. These lines represent most of the new lines under development in the corn breeding project.

b. Corn Rootworm. In tests conducted at Brookings, S. Dak., the following lines have showed superior tolerance to the corn rootworm: HD2187, B57, CI21E, Mo22, B14, N38A, N8B, SD10, Oh05, A251, CI38B, Zap15, Zap13, Mex6, LRC3,1, and LRC3,6. One of these lines will be released as soon as a special genetic stock for tolerance to rootworm larvae. Two synthetics have been initiated involving these lines. The performance of the most promising lines was relatively comparable under irrigation and dryland where there was a moisture stress.

The number of lines tested during 1967 and percent with an acceptable level of performance within each type of material tested follows: (a) inbreds 139 - 22%, (b) single crosses 267 - 24%, (c) $S_1 169 - 40\%$, (d) $S_2 362 - 49\%$, (e) $S_3 135 - 37\%$, (f) $S_4 21 - 33\%$, (g) synthetics 30 - 60%. The

increased number saved in later generations indicates the positive effect of selection in previous generations. The synthetics represent corn belt maturity in Central and South American lines where numerous species of rootworms are indigenous.

c. Miscellaneous Insects. Research being conducted in India under PL 480 has led to the development of artificial rearing techniques for two stalk borers, Chilo zonellus and Sesamia inferens. Maize germ plasm with resistance to C. zonellus has also been found.

2. Small Grain and Sorghum Insects

a. Greenbug. Studies at Brookings, S. Dak., showed that cummulative amounts of greenbug feeding produced a quantitative, detrimental decline in chlorophyll content and rate of photosynthesis, an increase in respiration, and little effect upon fresh weight, dry weight, and leaf area. Resistant barley leaves maintained a higher chlorophyll content. Soluble protein content of barley leaves was not significantly altered by greenbug feeding. Peroxidase activity increased markedly at the site of infestation in both susceptible and resistant varieties. Phenolic content of infested leaves was maintained at a higher level than in the control leaves. Malic dehydrogenase activity and isoenzyme patterns remained unchanged during greenbug feeding.

One hundred and twenty-six wheats from Argentina tested in Oklahoma for greenbug resistance were all susceptible to biotype B greenbug. Three thousand World Wheat Collection entries also were susceptible to the same biotype.

Will barley, which is resistant to both greenbug biotypes and to the corn leaf aphid, was found to be susceptible to the yellow sugarcane aphid, Sipha flava.

b. Wheat Stem Sawfly. Hybrid material containing P. I. 94585 (solid-stemmed) selections were tested in the field at Conrad, Mont., for resistance to sawfly attack. Some of them show promise of maintaining stem solidness stability (resistance) with desirable agronomic characters. The rescreening of Turkish varieties for new sources of resistant germ plasm was continued. Many of the resistant and intermediate varieties have been found to be late or hollow-stemmed.

At Minot, N. Dak., dyed plastic sheeting was used as light filters to determine associations between several wheat plant characters and solar radiations at specific wavelengths. In the hard red spring wheat variety, Fortuna, stem solidness (and resistance) was associated with amount of radiation at wavelengths of 575 and 700 millimicrons. Increased light at 575 mu increased solidness; increased light at 700 mu decreased solidness.

- c. Fall Armyworm and Corn Earworm. In preliminary sorghum insects resistance tests, 355 entries from India were evaluated for resistance with laboratory reared larvae. Ninety-three were resistant to the fall armyworm and 75 to the corn earworm.
- d. Rice Insects. At Crowley, La., over 1000 varieties, including entries of the Uniform Yield Nursery, Arkansas Disease Nursery, New International Blast Nursery, and World Collection of Rices were screened for the host plant resistance to the rice water weevil and stem borers. Twenty-seven of these varieties apparently had some degree of resistance to the rice water weevil.
- e. Cereal Leaf Beetle and Hessian Fly. New and superior sources of resistance to the cereal leaf beetle in spring wheats (C.I.11490, C.I.9321, and C.I.9294) and winter wheat (C.I.8519) are now being utilized to develop a new series of backcrosses with commercial spring and winter varieties. Resistant F₃ and F₄ lines from crosses of both winter and spring varieties with cereal leaf beetle resistant lines have been developed at Brookings, S. Dak., and tested at Galien, Mich.

At East Lansing, Mich., adequate sources of germ plasm with resistance to the cereal leaf beetle have been found in wheat. Research has been initiated at Brookings, S. Dak., to incorporate resistance to the cereal leaf beetle in both hard red spring and winter varieties of wheat. Successful crosses were made between lines rated resistant to the cereal leaf beetle and the spring varieties Chris and Crim and the winter varieties Hume, Winalta, and Pawnee. More than 200 F4 winter wheat progenies from the crosses of resistant lines to Hume (38), Winalta (129), and Pawnee (44) have been rated resistant.

The higher level of resistance recently identified in laboratory tests in the spring wheat lines C.I.11490, C.I.9294, and the winter wheat line C.I. 8519 has been incorporated into the breeding program.

Spring wheat resistant lines, on which young larvae have not been able to survive, have been crossed with Justin, Crim, Chris, and several other advanced hard red spring wheat lines.

At Lafayette, Ind., Purdue wheat breeders have used pubescent spring and winter wheats, Agropyron, and Triticum persicum fuliginosum as resistant parents crossed with well adapted Hessian fly resistant wheats. Second and third generation hybrids are now ready for selection.

No highly resistant oats have been found. Crosses that were made with lines showing promise of resistance earlier have not shown resistance in field tests. Many of these earlier lines with potential, C.I. 4042, C.I. 4051, C.I. 4706, and C.I. 7495, may have escaped infestation.

Three or four lines in barley appear to have some resistance to the cereal leaf beetle. C.I. 6469 appears to be the most resistant. It has been crossed

Forty entries of unknown resistance from Tennessee were selected from the breeding nursery and evaluated to identify their resistant genes. All resistant entries had the $\rm H_{2}$ resistant gene.

Thirty-five foreign wheat entries being used as parents in the hybrid wheat program were evaluated for Hessian fly resistance. One entry was resistant to Races A and B, indicating that it has either the $\rm H_5$, the $\rm H_6$, or an unknown resistant gene.

One hundred and seventy-two new wheat introductions were tested to Races A and B. Nine entries reacted resistant to Race A, seven of which also reacted resistant to Race B. These wheats will be added to the gene pool of germ plasm resistant to the Hessian fly.

Eight hundred and twenty-five entries of wheat crosses having agrotriticum parentage were evaluated for resistance to Race D. One hundred and sixteen reacted resistant and will be added to the resistant germ plasm gene pool.

At Manhattan, Kans., approximately 2,100 lines were tested to the Great Plains race in the greenhouse. Entries from the Kansas intra-state nursery contained selections have the Parker, Pawnee, and Warrior types of resistance combined with wheat streak mosaic resistance. The Kansas observation nursery contained entries having the Parker, Ottawa, and Pawnee types of resistance combined with Bison, giving good wheat streak mosaic resistance. The Kansas breeding nursery consisted of the wheat crosses Shawnee X Apache, Apache X Parker, and Scout X 12855.

Parker, a Hessian fly resistant wheat variety having Marquillo resistance, was released.

Two hundred entries with Monon (H_3) , Benhur (H_6) , and Stadler resistance were evaluated against the Great Plains race; 91 entries reacted resistant.

Three new Hessian fly resistant wheat varieties were released in 1967. Riley 67 and Arthur are Indiana releases and have the H3 resistant gene. Parker, a Kansas release, has the Marquillo resistant gene.

Genetic studies with F_2 and backcross generations indicate that the ability of Race A and Race E to infest varieties resistant to the Great Plains race is simply inherited, probably controlled by one or two recessive genes.

Retardation of cruxin activity is believed to be associated with plant stunting observed in plants attacked by Hessian fly larvae.

Studies conducted on the possible relationship between growth regulators and Hessian fly resistance in wheat showed that all the wheats tested infested or non-infested (Turkey, Monon, Knox 62, and Ribeiro) possessed growth regulators. Ribeiro was resistant to all races, and had fewer auxins than did the other varieties. The Hessian fly larvae removed more auxins

with the variety Dickson, and the progeny is in the F_3 generation. Resistant lines have also been crossed with the varieties Dickson, Parkland, and Trophy. Resistance to the cereal leaf beetle in wheats is related to the amount and type of leaf pubescence. Pubescence has a threefold effect on the beetle, acting as a deterrent to oviposition, promoting desiccation of eggs, and inhibiting first instar larval feeding.

Wheat breeding material from five State experiment stations in the eastern soft wheat region and three State experiment station in the hard red winter wheat region were evaluated at Lafayette, Ind., for Hessian fly and/or cereal leaf beetle resistance. Approximately eleven thousand lines, hybrids, varieties, and selections were evaluated for Hessian fly and/or cereal leaf beetle resistance.

Non-preference by cereal leaf beetle adults for oviposition on pubescent leaf surfaces continues to be the major mechanism of resistance in wheats. Antibiosis also occurred in the form of egg desiccation when eggs were placed on pubescent leaf surfaces, and very little larval weight gain occurred when larvae fed on pubescent leaves.

f. Hessian Fly. Thirty-six entries from Georgia Agricultural Experiment Station containing crosses between $\rm H_2$ resistant Georgia 1123 and $\rm H_6$ resistant Benbur and Knox 62 were evaluated for resistance to Races A and B. Many reacted resistant to Race A, but only one entry reacted resistant to Race B.

Approximately 6,000 head selections, hybrids, and lines from the Purdue regular fly nursery, preliminary yield nursery, advanced yield nursery, fly stem rust nursery, and Septoria nursery were evaluated for resistance to Races B and D in replicated tests. All wheats have one or more of the $\rm H_3$, $\rm H_5$, $\rm H_6$, or Marquillo resistant genes in their parentage. Many of the $\rm F_2$ and backcross lines containing the $\rm H_5$ gene in combination with one or more of the $\rm H_3$ or $\rm H_6$ genes were saved, vernalized to break the winter habit, and utilized in the crossing program in the greenhouse. In the fall nursery at New Carlisle, Ind., approximately 4000 entries of small grains were evaluated for cereal leaf beetle resistance. Entries included Purdue breeding material with Agropyron parentage, pubescent wheats, and $\rm F_2$ and $\rm F_3$ space planted wheats having pubescent Triticum persicum fuliginosum and pubescent spring and winter common wheats as resistant parents.

Arthur, a Hessian fly resistant wheat variety, was released by the Indiana experiment station in cooperation with the Entomology and Crops Research Divisions of the U. S. Department of Agriculture. Arthur is a very high yielding wheat having the $\rm H_3$ gene for resistance to Races A, C, and F.

Other material evaluated were as follows: One hundred and fifty advanced wheats from Ohio with Hessian fly resistance in their parentage were tested to Races A and B. Many resistant types occurred, all with the H3 gene involved.

from plants that were susceptible to the race than those that were resistant. Some growth inhibitors were identified: IBA (indolebutyric acid) was the major auxin removed by the larvae of Races A, B, C, and D; however, some IAA (indoleacetic acid) was also removed.

Hessian fly extracts of Race B and Race D larvae prevented or reduced IBA (synthetic growth producing auxin) and IAA (natural growth producing hormone) activity on wheat and oat coleoptiles, when tested in artificial media. Coleptiles did not elongate as much as when the growth hormone was used alone.

e. Miscellaneous Insects. Studies conducted in India on insect pests of sorghum and millet under PL 480 indicate that lines are being found with resistance to insects that can be used to develop varieties in India with resistance to several sorghum and millet insect pests.

G. Insect Vectors of Diseases (3.6 SMY)

1. Corn. A non-persistant virus found infecting field corn was proven to have physical-chemical and host range characteristics similar to those of bromegrass mosaic virus. Seedlings mechanically innoculated develop local lesions on innoculated leaves followed by systemic invasion, whorl necrosis and is ultimately lethal. Transmission trials with English grain aphid, green peach aphid, corn leaf sphid, oat-bird cherry aphid, greenbug, and by soil innoculations have been negative. Transmission has been successful with rootworm larvae and adults of D. virgifera and D. undecimpunctata howardi.

Trapping studies conducted under a grant at the University of Missouri, on insect transmission of viruses causing stunting of corn showed that at least 36 aphid species have been found in corn fields in the Missouri Area. Three known vectors have been trapped - corn leaf aphid, greenbug (maize dwarf mosaic), and brick-red sowthistle aphid (sugarcane mosaic virus).

Tests conducted under a cooperative agreement at the Ohio Agricultural Research and Development Center on the role of mites as vectors of corn viruses have produced some interesting data on aphid movement of mites.

Large numbers of eriophyid mites were found on grease slide traps at the time of seedling emergence (June 1). Numbers of mites moving in the air began to drop off July 3 when mites first began to appear on corn (Oh 28, B-37) inbreds. A steady drop in mite movement occurred from July 3 to August 21 while mite numbers of corn (especially B-37) increased.

2. Small Grain At Brookings, S. Dak., recovery of BYDV from field populations of Macrosiphum avenae, Rhopalosiphum maidis, R. padi, R. rufiabdominalis, and Schizaphis graminum showed that in years when aphid populations and incidence of BYD were low, viruliferous aphid populations were also low. Virus was also recovered from trapped migrating R. maidis alates.

Studies conducted at Brookings, S. Dak., also indicated that plant age at time of attack by barley yellow virus, influence the plant reaction obtained. When Selkirk (hard red spring) and Langdon (durum wheat) were infested at four different plant ages with 0, 2, 4, and 8 viruliferous aphids for three days highly significant differences resulted from infection with virus for components of yield including heads per plant, plant height, heading date, kernels per head, 1000-kernel weight, and weight of kernels per head. Highly significant differences due to age of plant at time of infection were recorded for tillering, heads per plant, height at 21 days and maturity, and heading data. Varieties responded differently for plant height, heading date, kernels per head, and 1000-kernel weight. Differences due to different number of viruliferous aphids were non-significant.

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FORAGE AND RANGE INSECTS Entomology Research Division, ARS

USDA and Cooperative Program

			: Scientist Man-Years F.Y. 1968 : Research Problem Area :										
Location of Intramural Work		1	_:	Fotal									
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Washington		:		:			0.1			:	0.1		
	Total Alfalfa	:		:	4.7	:	0.7	:		:	5.4		
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Intramural program is supplemented by extramural support representing (a) 0.7 SMY at State Agricultural Experiment Stations 1/2 and (b) PL 480 funds in 1 country representing 85,065 U.S. dollars equivalent.

^{1/} RPA 112 0.7.

Problems and Objectives

Numerous insect pests that attack forage and range plants in the United States lower seed production, reduce the yield and quality, and decrease the abundance of range plants. Certain insects are involved in the transmission of forage-crop diseases. These losses are estimated at more than \$350 million annually. Among the more important insect pests are grasshoppers, lygus and other plant bugs, stink bugs, seed chalcids, the alfalfa weevil, root borers, spittlebugs, leafhoppers, and a variety of aphids. Insecticides are used to control some of these insects but they are often costly and may create residue hazards in meat and milk as well as adversely affect wildlife.

Major objectives of the work are to develop new or improved control methods by utilization of:

- 1. Chemical control methods that will avoid harmful residues.
- 2. Biological control methods parasites, predators, and pathogens.
- 3. Germ plasm to develop insect resistant varieties.
- 4. Knowledge of insect biology and ecology to develop control measures.
- 5. Sex attractants, sterilization techniques, and other new approaches.

Progress - USDA and Cooperative Program

RPA 112 - RANGE MANAGEMENT

A. Basic Biology, Physiology, and Nutrition (0.9 SMY)

1. Grasshoppers. In central and southeastern Arizona studies on factors influencing grasshopper populations and plant damage were continued on several intermountain rangelands. 1968 spring vegetation was abundant and favorable for grasshopper survival and development, but spring temperatures were below normal and development was about two weeks later than in 1967. Spring populations on three study areas showed a slight increase and more variability in 1968 over 1967: 1968 populations per square yard ranged from 2.2 to 7.9 as compared to from 2.0 to 3.7 in 1967. Late summer populations on four study areas in 1967 differed little from those of 1966, ranging from 3.9 to 8.0 and 2.9 to 8.2 per square yard, respectively. In 1967 damage to forage on an area averaging 6.6 grasshoppers per square yard was approximately 85%. Damage in 1968 is expected to be less due to an abundance of weedy food plants.

Under a research grant to Kansas State University, it was observed that the species of grasshoppers in cultivated pastures appeared earlier and developed more rapidly than those in native grasslands. There were no significant changes in food plant preferences during the different life stages. Slant-faced grasshoppers (Acridinae) were generally grass feeders and their ingestion percentage often ranked with the abundance of grass species. The Oedipodinae (band-wings) were grass feeders or mixed feeders

preferring grasses. The spine-breasted grasshoppers (Cyrtacanthacridinae) were classed as forb feeders or mixed feeders preferring forbs.

A study was initiated under a cooperative agreement with the University of Idaho of the food habits of rangeland grasshoppers. The crop contents of more than 1,000 grasshoppers from different areas have been analyzed and coded for data processing.

B. Insecticidal and Cultural Control (1.5 SMY)

1. Grasshoppers. At Bozeman, Mont., 38 candidate insecticides were screened by topical application to Melanoplus sanguinipes. Compared to a malathion standard, ten experimental compounds were superior at equal dosages.

In small plot field tests Dursban was effective at 2 ownces per acre, and SD 8447 was promising at 8 ounces in comparison to malathion at 10 ownces per acre.

In repeated ULV applications by air to 80-acre plots of rangeland, carbaryl at 8 ounces actual per acre averaged 95.9% control with a sticker and 98.6% without a sticker. Azinphosmethyl applied at 3 to 6 ounces actual per acre gave controls ranging from 95 to 99%. Bay 39007 applied at 6 ounces actual per acre averaged 97.7% control. Pursban applied at 1 ounce actual per acre did not give satisfactory control. Technical malathion at 8 fluid ounces per acre gave 96.1% control.

C. Biological Control (1.5 SMY)

1. Crasshoppers. Studies on the pathogens of grasshoppers and their potential use in grasshopper control have involved 11 specific or groups of pathogens in the laboratory while field studies, both natural and applied, have involved a virus and a protozoan. Field studies have established that the incidence of disease can be increased significantly through the application of pathogens on grasshopper baits.

A study of the natural occurrence of Nosema locustae, a protozoan, in grasshoppers in Camas County, Idaho, is in the sixth year. It is providing information on natural spread of infections which will be useful in predicting the course of epizootics after disease applications.

At San Carlos, Ariz., in September 1967, 24.8% of the adults of Trachyrhachis mexicana were parasitized by the nemestrinid fly, Trichopsidea clausa. In three areas of extreme southern Arizona adults of Morseiella flaviventris and Boopedon nubilum were parasitized by the nemestrinid, Neorhynchocephalus sackenii, at rates of 10.3, 43.3, and 43.2%. Three hundred ninety-two larvae of N. sackenii were collected and redistributed at San Carlos. In 1968, parasitism of spring grasshoppers at San Carlos by flesh flies was less than 1%.

Research under a cooperative agreement with Montana State University showed that the composition of a polyhedral virus of grasshoppers was 80.2% protein, .9 to 1.0% deoxyribonucleic acid, 0.6 to 1.9% polysaccharide and the balance apparently contains lipid fractions. A crystalline virus of grasshoppers was less complex, containing about 67% protein and 29% ribonucleic acid.

Under a cooperative agreement with Montana State University six major classes of lipids were isolated from the total lipids of grasshoppers infected with Malamoeba locustae and disease free grasshoppers. Free fatty acids, sterols, diglycerides, wax esters, and polar lipids composed 25% of the total lipid and tryglyceride the remaining 75%. Myristic, palmitic, palmitoleic, stearic, oleic, and linolenic acids are the principal fatty acids. There was no appreciable quantitative or qualitative difference in the fatty acid composition of the fat bodies of infected or disease-free grasshoppers.

At Bozeman, Mont., the search for a suitable larval media for rearing parasitic flies of the genus Blaesoxipha has been unsuccessful to date, although animal liver formulations seem to be the most promising. Laboratory infections of American grasshoppers by the European Anthomyiid fly, Acridomyia sacharovi, have not been successful.

- D. Insect Sterility, Attractants, and Other New Approaches to Control (0.0 SMY)
- 1. Grasshoppers. Under PL 480 research at The Hebrew University, Jerusalem, Israel, broadcasting recorded masticating sounds of the desert locust to nymphs of the same species produced a weak but insignificant feeding response. This is contrary to previous results when a definite response occurred.
- E. Evaluation of Equipment for Insect Detection and Control. (0.3 SMY)
- l. Grasshoppers. Three types of spinner nozzles were compared at Bozeman, Mont., by droplet size and efficiency of control of grasshoppers on 160-acre rangeland plots. Technical malathion was applied by aircraft at 6 fluid ounces per acre. All nozzles gave highly satisfactory control. The Micronair, however, had the least portion of its volume (20%) in drops under 100 μ in diameter and showed the largest ground deposit as compared to the Fischer (EA-12) and turbaero (Bals spinner) atomizers. All airplane tests were in cooperation with Plant Pest Control Division.
- F. Varietal Evaluation for Insect Resistance (0.2 SMY)
- 1. Grasshoppers. Of 26 grass varieties tested for resistance to feeding by Melanoplus sanguinipes at Bozeman, Mont., six were non-preferred and resulted in less than normal weight gain by the grasshopper. They were Stipa viridula (green stipa), Agropyron dasystachyum (P-15581), Festuca ovina var. duriuscula (Durar), F. arundinacea (Kenmont), Dactylis glomerata (Pennlate), and Bromus inermis (Lincoln).

RPA 207 - CONTROL OF INSECT PESTS OF FIELD CROPS

A. Basic Biology, Physiology, and Nutrition (1.9 SMY)

l. Alfalfa

a. Alfalfa Weevil. In Maryland adults of the alfalfa weevil were much later moving into alfalfa fields in the fall of 1967 than in previous years. A similar pattern appeared to be true throughout many of the Eastern and Notheastern States. In both Maryland and New Jersey larval populations were much lower in the spring of 1968 and peak development delayed by 2-3 weeks.

Survival of alfalfa weevil adults, with the synthetic hormone, 10, 11 epoxfaresenic acid methyl ester, was good when alfalfa was supplied after treatment, but when treated weevils were placed on sugar water after treatment survival was very low. Tests with additional synthetic hormones show that some compounds required a much lower dosage to break diapause and initiate normal oviposition (10 µg/weevil as compared to the present 100 µg/weevil) and had additional effects such as a pronounced juvenilization of larvae and carryover of this effect in progeny of treated adults.

In the laboratory at Beltsville, Md., Sonora alfalfa, grown in flats and infested when 2-3 weeks old successfully supported populations of up to 500 alfalfa weevil larvae per flat. It was shown that 1000 adults per week could be produced by this method.

The addition of alfalfa pollen to cages containing both stems and leaves for food increased alfalfa weevil egg production by 35%. When weevils were confined with alfalfa stems only there was very little oviposition; the addition of pollen increased egg production by 96%. Parafilm "straws" with a diameter similar to alfalfa stems were 53% as effective as alfalfa stems and leaves.

Studies were conducted at Beltsville to determine the effect of rhythm and photoperiod on oviposition. A periodicity effect was not evident as no differences were detected in the total eggs laid by weevils when examined at 8 a.m. and at 8 p.m. Under a regime of 16 hours light and 8 hours darkness (16L 8D), significantly more eggs were laid than under 16D 8L. Significantly less eggs were laid under conditions of constant light (24L) and constant darkness (24D) than under 16D 8L. Also egg laying essentially ceased after 100 days under constant conditions but continued up to 120 days under the variable conditions.

The western strain of the alfalfa weevil has now reached Dawson County, one of Nebraska's prime alfalfa districts. Studies were initiated in 1967 to determine whether the weevil will develop in greater numbers and become more of a pest in this new location than it has been in other areas during its past history in Nebraska. Its seasonal occurrence generally followed

the same pattern as it does elsewhere in the United States. The rate of parasitism by Bathyplectes curculionis fluctuated from 6.1% to 53.5% during the period of weevil development and averaged 15% for the season.

- b. Spotted Alfalfa Aphid. At Lincoln, Nebr., significant differences were found between the sexual and asexual forms of the spotted alfalfa aphid in regard to adult weights, post reproductive period, nymphs produced per day, total number of nymphs produced, and total length of life. The reproductive capacity and length of life was greater among the asexual biotype.
- c. Pea Aphid. At Tucson, Ariz., four new biotypes of the pea aphid were differentiated by studying populations from Bakersfield, Calif., (BPA); Corvallis, Oreg., (CPA); Mesa, Ariz., (MPA); Reno, Nev., (RPA) at 60, 68, 76, and 84° F in temperature controlled chambers. The longest nymphal stage at 60° was demonstrated by the MPA biotype followed by CPA at 68, 76, and 85°. The shortest nymphal stage at 60° was shown by the CPA biotype followed by BPA at 68 and 76°, and RPA at 84°. Additional evidence for separating the biotypes was obtained when the four populations caused differential seedling survival among four pea aphid resistant alfalfas. Highly significant differences in seedling survival and mean seedling height were obtained among varieties and aphid biotypes.

At Tucson, Ariz., fecundity of the pea aphid decreased when aphids were transferred from broadbean to alfalfa and increased when transferred from alfalfa to broadbean, irrespective of the number of days exposed to the initial host. The most significant changes occurred when aphids were confined to initial hosts for very short (5-10 days) or very long (40-80 days) periods.

d. Lygus. At Tucson adult longevity of lygus bugs was markedly reduced when reared on fresh beans without sugar supplements. However, there was no statistical difference in egg deposition, hatching of nymphs, or nymphal mortality when bugs were reared on beans treated with solutions of 10% sucrose, 10% honey, or tap water. The length of the life cycle was similar between bugs reared on fresh beans treated with 10% sucrose and those treated with tap water.

At Mesa, Ariz., weekly lygus bug surveys in alfalfa and cotton fields showed no nymphs and very low adult populations in cotton fields. Populations were higher in alfalfa fields remote from cotton than in fields adjacent to cotton. Highest populations were observed in alfalfa in August. The data suggest that very little migration of lygus from alfalfa to cotton occurred during the period July 1, 1967, to June 30, 1968.

Under a grant to the University of California at Davis, an enzyme, endopolygalactusonase, produced in the salivary gland of lygus bugs was indicated as a primary cause of the plant damage which results from lygus bug feeding. At dilutions of 1:500,000 the enzyme digested more than 5 mg of plant tissue per hour. Extracts from homogenized whole bugs on the

salivary glands were active but extracts from bugs from which the salivary glands were removed showed no activity. Females showed, on the average, twice the activity of males.

e. Seed Chalcid. At Mesa, Ariz., field populations of the alfalfa seed chalcid entered initial diapause in late August and reached full diapause during the last week in November. Termination of diapause entered initial stages in the middle of April, peaked about the middle of May, and receded during middle of June. There was some evidence that a very small percentage of the population remained in diapause more than one year.

In studies under a grant to the University of Wyoming, attempts to induce the alfalfa seed chalcid to oviposit in several artificial media have been unsuccessful. Treating the media with extracts of different chemicals from alfalfa had little effect on ovipositional response.

f. Aphids. At Las Cruces, N. Mex., research under a grant to New Mexico State University to develop chemically defined diets for aphids showed that a temperature of 69° F was optimum for rearing the cotton aphid. Optimum concentrations of zinc in the diet were 0.0001 and of iron 0.001%. Manganese was detrimental at all concentrations. Omission of some salt mixtures resulted in an increase in growth or longevity. Calcium showed a synergistic effect with one or more elements in the salt solution but little effect in the absence of salt mixtures.

2. Clover

- a. Sweetclover Root Borer. Field observations on Walshia micecolorella, sweetclover root borer, indicate that it completes but one generation per year in the vicinity of Lincoln, Nebr. As during the previous year, the insect was abundant only in sweetclover nurseries where plantings have been made, in close proximity to each other, each year for several years. It appears that this concentration and continuation of sweetclover may be an important factor in building up infestations.
- b. Sweetclover Weevil. Studies at Lincoln, Nebr., in 1966 indicated that coumarin may be a natural attractant for the sweetclover weevil. However, additional studies using coumarin baited traps produced erratic results indicating that while coumarin may show some attractiveness, it probably is not solely or primarily responsible for attracting weevils to sweet-clover.

3. Grass

a. Bromegrass Seed Midge. At Lincoln, Nebr., larvae of a bromegrass seed midge entered diapause inside the florets about the time the grass seeds matured in early July. Laboratory studies indicated that this diapause cannot be broken, even after exposure to low temperatures, until the larvae are subjected to approximately the same mositure conditions which

initiate seed germination. Adult emergence will take place in approximately 10-14 days after the initial application of the moisture.

- b. Billbugs. At Corvallis, Oreg., adults of the billbug, Sphenophorus venatus confluens overwintered in plant debris in or near orchardgrass fields. The billbugs fed in April, mated and oviposited eggs throughout May. Eggs were laid inside the stems or near a leaf sheath. Larvae usually fed inside the stem until larval growth became restricted by the size of the stem. Larvae that hatched from eggs deposited on the leaves fed briefly then moved to the roots where they completed their development. Pupae were first found in early August. A few teneral adults were observed in mid-August which marked the beginning of emergence of new adults. Newly emerged adults remained inactive for 6 to 14 days before searching for a suitable overwintering site.
- c. Sod webworm. At Corvallis, Oreg., 60% of the sod webworm, Crambus trisectus, larvae reared on Shorey's artificial diet, reached the pupal stage. Males and females laid fertile eggs. Chewings fescue leaves were superior to roots as larval food for C. trisectus. After 25 days, 95% of the leaf-fed larvae were alive compared to 40% of the root-fed larvae. The average weight of root-fed larvae was 15 mg, where, for leaf-fed larvae it was 62 mg. Mortality was 35% for individually reared larvae and 56% for group-reared larvae. No cannibalistic activity was observed.

In other experiments at Corvallis, additives were highly effective in reducing growth of microorganisms in sod-webworm cultures with no adverse affects on larvae. The most effective additive was a mixture of 1.25 gm methyl p-hydroxybenzoate, 0.63 gm sorbic acid, and 1.25 ml of 40% formaldehyde in 400 ml of water.

At Corvallis, Oreg., first instar larvae of C. leachellus cypridalis and C. trisectus were reared at 24° C - 16 hour and 24° C - 10 hour photophases. After 48 days larvae of both species in the 16 hour group developed without interruption. In the 10 hour photophase all trisectus larvae entered diapause, but reaction of cypridalis was variable. Induction of diapause in cypridalis was clearly dependent on an interaction of temperature and photoperiod but not in the case of trisectus where photoperiod acted independently of temperature. Photoperiod was the key factor in termination of diapause of two sod-webworm species. Diapausing larvae were exposed to 24° C - 10 hour and 24° C - 16 hour photophases. In the short day treatment the trisectus larvae were still in diapause after 50 days, but all cypridalis larvae terminated diapause after 37 days although larval growth was retarded. In the long day treatment both species terminated diapause after 24 days. Clearly, the short photo-period exerted a strong influence on breaking diapause in cypridalis although the photo-period effect diminished with prolonged exposure to temperatures favorable for growth.

B. Insecticidal and Cultural Control (1.2 SMY)

1. Alfalfa

a. Alfalfa Weevil. At Beltsville, Md., laboratory screening 9 out of 17 experimental insecticides were promising to control the alfalfa weevil. Encapsulated phorate and parathion were compared with granular formulations at 1 and 2 pounds per acre. Adult weevils were exposed in a chamber for 4 hours every 5 days until mortality dropped below 50% or until 40 days elapsed. Phorate capsules and granules at both rates remained effective through this period (80% mortality or higher) while parathion capsules and granules at the 1 pound rate were ineffective after 20 days and only moderately active at 40 days at the 2 pound rate.

Topical treatment of larvae and adults of the alfalfa weevil with the same insecticides showed that there was a highly significant correlation in response at the LD-95, and a positive but not significant correlation at the LD-50. This indicates that adults, which are much easier to store and handle, can be used in the laboratory as an indication of larval insecticidal activity in the field. There was no difference found in the response of field and first generation laboratory reared adults to topical treatments of three insecticides.

Phorate, carbofuran, and Dursban were applied on March 26 in granula; form at 1 and 2 pounds per acre for alfalfa weevil control. A second application of carbofuran and Dursban was made to the same plots on April 9. Larval feeding damage was evaluated on May 8. Carbofuran at the 2 pound rate gave very effective control at both application dates. Carbofuran and phorate gave good control at the rates and dates applied. Dursban did not effectively control the weevil in these tests.

Sprays of carbofuran, Geigy GS-13005, and phorate at 1, 1, and 1/2 pound per acre respectively, were more effective against the alfalfa weevil than methoxychlor, azinphosmethyl, methyl parathion, or a mixture of malathion and methoxychlor at recommended rates.

Eight insecticides were applied at comparable dosages to small plots of alfalfa at Beltsville. Fifteen days after treatment carbofuran, Bay 39007, and Union Carbide UC-34096 gave very effective control of the alfalfa weevil.

b. Army Cutworm. At Lincoln, Nebr., sprays of diazinon at 1 and 1/2 round, carbaryl at 1-1/2 pound, and trichlorfon at 1 pound per acre applied to alfalfa, significantly reduced the population of army cutworm. Trichlorfon at 1/2 pound and carbaryl at 1 pound per acre were not effective. However, the best treatments only effected a 66% reduction and no differences in alfalfa growth was observed.

2. Grass

- a. Billbugs. Post-harvest field burning was ineffective in reducing billbug populations in orchardgrass in tests at Corvallis, Oreg. A total of 409 larvae was found in pre-burning samples, whereas, 383 larvae were found in post-burning samples. Among larvae found in post-burning samples, only five were injured by burning.
- C. Biological Control (0.9 SMY)

l. Alfalfa

a. Alfalfa Weevil. Tetrastichus incertus was recovered from alfalfa weevil larvae collected in Crawford, Mercer, and Washington Counties, Pa., and in Highland and Pulaski Counties, Va. The parasite was recovered in only 2 of the 10 New Jersey survey fields add did not exceed 2% parasitism. Six of the 10 fields in Maryland showed the presence of the parasite; an increase over last year. The highest incidence of parasitism for a single Maryland field was 24%.

Bathyplectes curculionis was recovered from 8 of the 10 Maryland survey fields. One field showed a peak parasitism of 60% and an average parasitism of 49% over a 2-1/2-month period starting April 18. This high level of parasitism occurred at the same time as maximum larval development this year. This syncronization of parasite and host populations appeared to be due to the delayed development of alfalfa weevil larvae due to cool temperatures. In New Jersey B. curculionis was found in all 10 fields with a peak of 31% parasitism in one field.

- b. Egyptian Alfalfa Weevil. At Mesa, Ariz., no recoveries were obtained of the egg parasite, Peridesmia discus or the larval parasite, T. incertus of the Egyptian alfalfa weevil during the spring survey of 1968, one year after release of the parasites.
- c. Pea Aphid. Research under a grant to the University of Kentucky showed that the parasite, Aphidius smithi, is a significant factor in the control of the pea aphid. A single female A. smithi exposed to 50 and 100 aphids parasitized 85 and 92%, respectively.

2. Clover

a. Aphids. A parasite, Aphelinus semiflavus, previously unknown in Nebraska, was detected in 1965, 1966, and 1967, but only when greenhouse cultures of the sweetclover aphid were started from field collected aphids in the fall. Aphid mummies caused by two other species (Praon palitans and Trioxys utilis) are occasionally detected in the field while mummies of Aphelinus are not. Natural field parasitism of the sweetclover aphid and the spotted alfalfa aphid by Λ. semiflavus varied from 4 to 24% as determined by laboratory rearing of field collected aphids. In laboratory

rearing studies A. semiflavus developed from egg to mummy in 5-8 days (temperature 22° C at night, 31° C day) and required another 6-9 days in the cocoon before adult emergence. The parasite developed equally well on three species of Therioaphis.

3. Grass

- a. White Grub. In 1962 field plots in Nebraska were treated with milky disease spores (a mixture of Bacillus popilliae and B. lentimorbus) for control of the white grub, Phyllophaga anxia. Although B. popilliae attacked the grub in the laboratory, subsequent field infection had not been noted until one of these plots was visited, with state cooperators, in the fall of 1967. At that time several grubs with typical milky disease symptoms were found in a meadow adjacent to the one containing the treatments. Further laboratory investigation in the Department of Entomology, University of Nebraska, demonstrated the presence of spores of B. popilliae in the blood of these grubs. This observation indicates the establishment and spread of B. popilliae under field conditions.
- b. Rhodesgrass Scale. Research at Weslaco, Tex., under a contract with Texas AGM University showed that aircraft releases of adult and preimaginal stages of Neodusmetia sangwani, a parasite of the Rhodesgrass scale, resulted in 50.0 and 47.8 establishment, respectively. A greater number of established colonies was found with air drops at 1 x 2 mile than 1 x 1 mile release densities.
- D. Insect Sterility, Attractants, and Other New Approaches to Control. (0.0 SNY)

1. Alfalfa

- a. Alfalfa Weevil. Research under a grant to Virginia Polytechnic Institute, Blacksburg, Va., showed that extracts from alfalfa using either a 1:1 acetone distilled water mixture or distilled water alone elicited an attractant or arrestant response from the alfalfa weevil. Both extracts were deactivated by heat or when left overmight in an uncovered dish at 3-5° C. When stored at this temperature in a covered container activity was retained for 12-15 days.
- E. Evaluation of Equipment for Insect Detection and Control (0.1 SMY)

1. Alfalfa

a. Alfalfa Weevil. At Beltsville, Md., flaming of dormant alfalfa to control the alfalfa weevil was conducted in the fall and spring. Evaluations on April 30, indicated that both fall and spring flaming and a combination of the two gave effective control up to that time. However, weevil development was delayed this spring, and evaluations on May 2 showed that high numbers of eggs were present and a late infestation could be

expected. Stubble treatment after early harvest both with an insecticide and by flaming did not give effective control on the second crop. Plots harvested on May 13 required an insecticide spray prior to harvest to adequately control the weevil, suggesting that under conditions more favorable to weevil development flaming would have to be supplemented with an insecticide treatment.

F. Varietal Evaluation for Insect Resistance (4.3 SMY)

1. Alfalfa

a. Alfalfa Weevil. At Beltsville, Md., laboratory screening and selection for resistance to the alfalfa weevil were concentrated on seven populations which previously had shown the highest level and frequency of resistant selections, and/or with good agronomic characteristics. Under laboratory conditions significant progress has been made for resistance to larval survival and development and adult feeding preference.

Small plot and nursery studies with the most promising laboratory selections in the 1966-67 program gave very promising results in the spring of 1968 both at Raleigh, N. C., and at Beltsville, Md. In the management trials the alfalfa weevil resistant selections yielded as high or higher than the commercial varieties Cherokee, Iroquois, Saranac, and Vernal. There were significantly lower numbers of larvae collected from the resistant selections MS Hp5 and MS Hp 6 than from other entries.

In the spring of 1968 seed of AWPx₃ was made available to alfalfa breeders as a source of alfalfa weevil resistant germplasm. This material was selected for resistance to larval development, adult feeding, and oviposition in laboratory tests.

Large cage field studies were established in July 1967 to compare the response of the alfalfa weevil to four alfalfa entries selected for resistance. Promising results were obtained with two entries selected for larval and/or adult feeding resistance. Entries selected primarily for oviposition resistance in the laboratory failed to express adequate resistance under these conditions.

The response of larvae from eight populations of the alfalfa weevil to alfalfa clones selected for resistance to Maryland weevils was compared in the laboratory at Beltsville, Md. Adults collected from the field in California, Utah, Montana, North Carolina, Ohio, Maryland, Pennsylvania, and New York were parents of the larvae used in the study. There was no significant difference in the response of weevil populations from the eight locations under conditions of this test. Larvae of all populations of the eastern strain developed faster than those of the western strain. Larvae from North Carolina and New York developed faster than those from Ohio, Pennsylvania, and Maryland.

- b. Egyptian Alfalfa Weevil. In field plots at Mesa, Ariz., 50 plants representing four genetic sources were selected free of damage under moderate to heavy Egyptian alfalfa weevil infestation. All plants have been placed in an isolated block for production of seed and recurrent selection tests.
- c. Aphids. At Tucson, Ariz., progenies of 11 pea aphid resistant experimentals and varieties of alfalfa were evaluated for resistance to biotypes ENT-A and ENT-B of the spotted alfalfa aphid. Seedling survival ranged from 94% for Syn. BB to 59% for Syn. EE in tests against ENT-A. Washoe and Dawson had 82 and 80% seedling survival, respectively. In tests against ENT-B, seedling survival ranged from 95% for Syn. BB to 73% for NS-39. Washoe and Dawson had 84% seedling survival.

At Tucson progenies of new Nevada and Utah experimental alfalfa were evaluated for resistance to ENT-A. The experimentals were developed for resistance to diseases, nematodes, and the spotted alfalfa aphid. Four of the six experimentals from Nevada (MSE-5, MSF-6, MSF-6, MSF-6) had resistance equal to the resistant check. Seedling survival ranged from 90-99%. Two Utah experimentals (U-5156, U-5157) had only moderate resistance.

At Mesa over 500 plants representing eight different genetic sources were screened for antibiosis to ENT-A, and 110 showed high resistance, 120 moderate resistance, 20 low resistance, and 250 were classed as susceptible.

In tests at Tucson 15 superior plants from a 24-clone polycross combination of Chilean alfalfa were selected for high level of resistance to ENT-A of the spotted alfalfa aphid. An additional 17 plants from Hairy Peruvian believed to have tolerance to lygus bugs were selected for high resistance to ENT-A. Thirteen progenies of Sonora parentage were selected from a new two-clone, high forage producing combination for resistance to ENT-A.

At Tucson 53 superior alfalfa plants were selected from a number of 2-clone combinations for combined resistance to ENT-A of the spotted alfalfa aphid and the Bakersfield, Calif., and Mesa, Ariz., biotypes of the pea aphid. All 2-clone combinations were developed in Arizona from selections out of Mesa-Sirsa alfalfa. Several plants were also selected for combined aphid resistance from two new experimentals developed in California.

In greenhouse tests at Tucson antibiosis reaction of pea aphid resistant alfalfa plants was non-significant between populations of the pea aphid reared on broadbean and alfalfa. Populations of pea aphids (Bakersfield, Calif., biotype) reared separately on alfalfa and broadbean in the greenhouse were held on susceptible and pea aphid resistant alfalfa plants for 10 days. Reproduction and survival of the original test population averaged 1.0 and 2.8 aphids per resistant plant from the population reared on alfalfa and broadbean, respectively. On susceptible alfalfa plants reproduction averaged 75 and 48 per plant from the population reared on alfalfa and broadbean, respectively.

A study was initiated at Lincoln, Nebr., to determine the effects of feeding by the pea aphid and the spotted alfalfa aphid on the yield and quality of three alfalfa varieties and one experimental synthetic. The alfalfas used were Vernal and Ranger which are susceptible to both aphids. and Dawson and Kansas State-10 which are resistant to both aphids. Three cages were each planted to four replications of the four entries. One was infested with pea aphids, the second with spotted alfalfa aphids, and the third was kept free of insects. In general, alfalfa infested by either aphid contained less carotene, protein, and digestible dry matter and more fiber than noninfested alfalfas. The resistant alfalfas in the pea aphid cage supported fewer aphids, produced more forage and contained more carotene than the susceptible alfalfas. Fiber, protein, and digestible dry-matter contents of the four alfalfas were not significantly different. The resistant alfalfas in the spotted aphid cage supported fewer aphids. produced more forage, contained less fiber and more carotene and protein than the susceptible alfalfas. Digestible dry-matter contents of the four alfalfas were not significantly different.

d. Lygus. At Mesa, Ariz., 3000 plants representing 32 different genetic sources were screened for resistance to field populations of Lygus. Twenty-five plants were selected free of lygus bug injury.

At Tucson, Ariz., 16 ratios (1:2, 3:2, 5:2, 7:2, 1:4, 3:4, 5:4, 7:4, 1:6, 3:6, 5:6, 7:6, 1:8, 3:8, 5:8, 7:8) of number of lygus nymphs to alfalfa buds were used to determine the ratio that gave normal development and survival of nymphs. Utilizing lygus susceptible alfalfa plants, ratios of 1:8, 1:6, and 1:4 gave zero, 25 and 25% mortality of nymphs, respectively, the lowest among all ratios tested. The 7:2 ratio gave 100% nymphal mortality. A ratio of one nymph to eight buds was used in testing 30 selections for lygus resistance. Five plants showed evidence of resistance when several nymphs died after a 14-day testing period, whereas, all nymphs on susceptible check plants survived and matured.

Pesearch at Manhattan, Kans., under a grant to Kansas State University, has shown that seedling plants of alfalfa varieties can be used to test for resistance to lygus bugs. Seedlings in the cotyledon stage were killed more readily than those in the unifoliolate stage. Plant damage was greater under 16 hours of light than under 0, 8, or 24 hours. Late instar lygus bug nymphs caused more damage than an equal number of adults. The synthetic alfalfa variety KS-10 that has resistance to the pea aphid and spotted alfalfa aphid was more resistant to lygus bugs than other varieties tested.

e. Seed Chalcid. At Mesa, Ariz., a significant reduction in alfalfa seed chalcid populations was found in all new synthetic alfalfas developed for resistance to this insect. Populations were eight times greater on susceptible check entries. A significantly fewer number of diapausing larvae was found in all resistant experimentals, suggesting that resistance either prevents initiation of diapause or terminates it prematurely.

f. Miscellaneous Insects. At University Park, Pa., the third cycle of selection of alfalfa for resistance to spittlebug and the potato leafhopper was completed this spring and compared with previous cycles. There was a trend toward increased resistance to spittlebug and to wilting leaflets caused by potato leafhopper.

Suudies were made at Lincoln, Nebr., to determine the relationships among leafhopper feeding injury, yield, and quality of alfalfa clones varying in reaction to this insect. Two field cages were each planted to three replications of 12 alfalfa clones. One cage was infested with 20 adult leafhoppers per plant, and the other was kept free of insects. In general, clones in the leafhopper-infested cage, regardless of visual damage score, produced less forage, contained less moisture, carotene, fiber, and protein than the same clones grown in the noninfested cage. The mean percent reduction from the noninfested controls for each of these characters, with the exception of fiber content, was less for the more resistant clones than the more susceptible clones.

At Tucson, Ariz., over 1600 plants were screened for resistance to root, stem, and leaf diseases. Nearly 250 plants were selected from this group for superior agronomic characteristics. Seed of this group was produced and the progeny screened for resistance to leafhoppers, Empoasca sp., and the spotted alfalfa aphid, resulting in the selection of 12 superior plants.

At Manhattan, Kans., under a contract with Kansas State University, a method was developed for screening alfalfa in the unifoliolate stage for resistance to the potato leafhopper. Among 28 varieties and four experimental alfalfas the winter hardy and variegated floral types were more resistant. One synthetic variety with resistance to the pea aphid, spotted alfalfa aphid, and with some resistance to the potato leafhopper has been approved for seed increase.

At Reno, Nev., under a cooperative agreement with the University of Nevada attempts, using thin layer chromatography, to identify the compounds in steam distillates of alfalfa that may be related to aphid resistance have been disappointing. When put through a charcoal column, the distillate loses most of its activity indicating that these compounds are probably bound to the charcoal. Attempts are underway to elute them from the charcoal.

Under a contract with the Research Triangle Institute, Research Triangle Park, N. C., a satisfactory technique, using thin layer chromatography, has been developed to analyze and separate into individual components the saponins in insect-resistant and susceptible varieties of alfalfa.

At Raleigh, N. C., under a cooperative agreement with North Carolina State University, several extracts from Lahontan alfalfa containing saponins were evaluated and two deterred feeding by the alfalfa weevil. Three saponin fractions from DuPuits alfalfa were tested and one showed deterrent activity. the others showed some attractancy at high concentrations.

2. Clover

- a. Sweetclover Root Borer. Walshia miscecolorella was found to infest all of the biennial species and varieties of sweetclover observed at Lincoln, Nebr. While some differences seem to exist between varieties and lines in regard to borer infestations, further work is needed to determine the nature and importance of the differences.
- b. Sweetclover Weevil. A feeding deterrent in sweetclover which appears to be primarily responsible for the resistance of Melilotus infesta to the sweetclover weevil has been isolated in pure crystalline form. It has been identified by physical and chemical methods as ammonium nitrate. Ammonium nitrate has been shown to act as a strong feeding deterrent for the weevil. This research was done under a grant to the University of Nebraska, Lincoln, Nebr.

3. Grass

a. Fall Armyworm. At Tifton, Ga., 441 bermudagrass clones were screened for resistance to first instar fall armyworm larvae, and 12 showed nonpreference.

At Tifton 1436 Pearl millet inbreds were screened as seedlings for resistance to first instar fall armyworm larvae. Approximately 4% were rated resistant, 28% intermediate, and 68% susceptible to larval feeding.

At Tifton, various lines of Pearl millet, peanut foliage, and Coastal bermudagrass with known resistance ratings to fall armyworms were analyzed for 6-MBOA (and BOA) content as possible indicators of resistance. The SPF data could not be related to the resistance ratings and since the 6-MBOA and BOA content, if any, is very low, these compounds appear not to be indicators of fall armyworm resistance in these crops.

b. Two-Lined Spittlebug. A highly variable collection of 404 clones of bermudagrass was screened at Tifton, Ga., for resistance to the two-lined spittlebug. Five percent were rated resistant, 47% intermediate, and 48% susceptible.

G. Insect Vectors of Disease (0.3 SMY)

At University Park, Pa., the common fungus fly, Bradysia sp., infesting greenhouses injured red clover and alfalfa seedlings and predisposed these plants to Fusarium roseum, and F. oxysporum f. sp. medicaginis, respectively. Control of Bradysia is essential in conducting insect-root rot investigations.

A study was made at University Park, Pa., to determine the relationship of Sitona hispidula, Fusarium, and bacterial wilt on alfalfa. S. hispidula reduced the growth of alfalfa and provided an entrance for Fusarium. A clear relationship was not established between S. hispidula and bacterial wilt.

RPA 701 - INSURE FOOD PRODUCTS FREE FROM TOXIC RESIDUES FROM AGRICULTURAL SOURCES

A. Insecticide Residue Determinations (1.8 SMY)

At Tifton, Ga., Imidan residues disappeared on corn in 7 days to 3.37, 3.68, and 5.78%; on soybeans in 15 days to 0.32, 0.50, and 0.92%; and on Coastal bermudagrass in 15 days to 1:06, 1.33, and 1.07% of the residues detected after spray applications at rates of 4, 8, and 16 ounces per acre. Residues of Imidoxon after 7 days on corn and 15 days on soybeans and Coastal bermudagrass were not detected.

At Tifton, Dursban was applied at dosages up to one pound per acre to corn in the field. The residues declined rapidly in the field but remained rather constant in silage. Samples taken on the day of treatment with 1 pound per acre averaged 15 ppm of residue. On the following day, Dursban residues in chopped corn taken just prior to ensiling had decreased to about 4 ppm. During 140 days of storage in the silo the residues declined to about 2 ppm. Residues of Dursban in corn and corn silage treated at 4 and 8 ounces per acre were proportionally lower. No residues of the oxygen analog of Dursban were found in any of the samples. Aqueous seepage from the silos contained traces of Dursban. No residues of Dursban or its oxygen analog were detected in the milk from cows fed the corn silage or in their urine. Their feces contained up to 0.31 ppm of Dursban but no detectable oxygen analog. Cholinesterase activity of blood from the animals fed the treated silage appeared to be unaffected.

At Tifton plots of Coastal bermudagrass and corn treated with fenthion emulsifiable concentrate at 0.5, 1.0, and 2.0 pound per acre were sampled immediately after treatment and at 1, 2, 7, 14, and 21 days posttreatment. Residue analyses for fenthion and five of its metabolites (oxidation products) were performed. Extensive oxidation of fenthion to the metabolites (primarily to fenthion sulfoxide) occurred almost immediately in the field. Residues of the six compounds were detected in the grass and all but the fenthion oxygen analog were found in the corn after various intervals of weathering in the field. Residues of fenthion and the metabolites in corn ensiled in gallon jars were persistant and oxidized at a lower rate than in the field.

At Tifton biological assays to determine the relative toxicity of fenthion and five metabolites were performed using first instar fall armyworm larvae. In order of decreasing toxicity the compounds were: fenthion 0-analog (P=0,S), fenthion (P=S,S), fenthion sulfoxide (P=S,S0), fenthion 0-analog sulfone (P=0,S0₂), fenthion sulfone (P=S,S0₂), and fenthion 0-analog sulfoxide (P=0,S0).

At Tifton the feces from beef steers fed silage treated with Bidrin (15.7 ppm dry basis) for seven days contained no detectable residues of Bidrin or Azodrin (3-hydroxy-N-methyl-cis-crotonamide dimethyl phosphate), a

metabolite of Bidrin. Urine samples taken concurrently with feces contained no detectable residues of Bidrin but Azodrin residues ranged from 0.36 to 0.51 ppm.

Residue analyses were made at Tifton of milk and feces from cows fed silage containing 13 or 23 ppm (wet basis) of Gardona. The feces contained 0.17 ppm or less (wet basis) of residue. No residues were detected in the milk.

At Beltsville, Md., pigs were fed alfalfa hay containing 34 ppm of DDT, 28 ppm of heptachlor epoxide, or 150 ppm of malathion. Samples of blood were taken at 2-week intervals throughout the feeding period. The maximum residues found were about 0.01 ppm of heptachlor epoxide, and 0.56 ppm of p,p'DDT. No malathion or maloxon were found.

At Yakima, Wash., a 10% granular formulation of Union Carbide UC-21149 was applied to the soil of potted alfalfa plants and field plots at 1/2, 1, and 2 pounds per acre of active ingredient. The greenhouse alfalfa was sampled 24 to 70 days after treatment and the field alfalfa was sampled seven days after treatment. No UC-21149 was found in any of the samples. UC-21149 sulfoxide and sulfone residues were found in amounts up to 5.7 ppm in the greenhouse alfalfa and up to 0.87 ppm in the field alfalfa. A 10% granular formulation was applied to the soil of potted alfalfa plants at 1, 2, and 3 pounds per acre. Analysis of the alfalfa 51 days after treatment showed no UC-21149 residues, but the sulfoxide ranged from 2 to to 14 ppm and the sulfone from 2 to 21 ppm.

Analysis at Yakima of rangegrass treated in Wyoming with ULV applications of Bidrin at 2 and 4 ounces per acre, showed residues of 13 and 51 ppm of Bidrin, respectively, four hours after application. In 45 days these decreased to 0.26 and 0.70 ppm.

At Yakima analyses of rangegrass treated with ULV applications of Bay 39007 at 6 ounces per acre showed 114 ppm on the grass immediately after treatment, 28 ppm after 7 days, and 6 ppm after 14 days. Azinphosmethyl applied at 4 ounces per acre showed 92 ppm immediately after treatment, 106 ppm after three days, 82 after seven days, and 47 after 14 days.

RPA 906 - CULTURE AND PROTECTION OF ORNAMENTALS AND TURF

A. Basic Biology, Physiology, and Nutrition (0.7 SMY)

1. White-fringed Beetles. At Gulfport, Miss., visible increases in larval sizes were noted for white-fringed beetle larvae fed different diets, but larval development beyond the second instar did not occur with any diet tested. The use of 0.5% methyl-p-hydroxbenzoate appreciably retarded microbial growth for upwards of 40 days with no detrimental effect on the larvae.

Viability of white-fringed beetle eggs from 1 to 38 days old and stored under different conditions was studied at Gulfport, Miss. Sixty percent of the eggs from 17 to 38 days old held at 92% RH and 55° F hatched after five months storage.

White-fringed beetle eggs of different ages were exposed to temperatures of 8°, 20°, and 30° F for 1 to 14 days. Eggs exposed to 8° F for one day failed to hatch. At 20° F eggs 3 to 10 and 8 to 15 days old hatched after exposures of one and three days, respectively. Eggs 21 to 28 days old failed to hatch when exposed to 20° F. for one day. Eggs 8-15 and 21-28 days old hatched after 14 days' exposure to 35° F.

An artificial rearing method using a potato-soil medium was developed for the white-fringed beetle under a research grant to Auburn University, Auburn, Ala. The length of the larval period was 5.5 months compared with 10.7 months in the field. Survival to the adult stage as high as 50% has been achieved. Conventional artificial diets tested have been unsatisfactory as the antimicrobial agents used were lethal to the larvae.

B. Insecticidal and Cultural Control (1.3 SMY)

1. White-fringed Beetles. At Gulfport, Niss., 32 compounds were tested as soil insecticides against newly hatched white-fringed beetle larvae. At 5 and 10 pounds per acre Dursban and Stauffer N-4446 (ENT 27045) gave 100% control. Thirteen other materials gave 100% mortality at the 10-pound rate.

Potting soil was treated with 10% granular disulfoton at the rates of 1, 5, 10, and 25 pounds per acre. Peanuts were planted and adult white-fringed beetles introduced. The 25-pound rate (2.5 pounds actual) gave 100% mortality for 10 weeks. The 10-pound rate (1.0 actual) gave 100% control for seven weeks.

White-fringed beetle larvae collected in a treated nursery showed the presence of dieldrin and DDT when analyzed by gas chromatography. Soil from the same locations also showed the presence of dieldrin and DDT. Biological tests indicated that the progeny of larvae collected from the same sites were resistant to dieldrin. Larvae from untreated sites showed no trace of insecticides and larvae reared from adults collected from the same sites showed no apparent resistance.

- C. Insect Sterility, Attractants, and Other New Approaches to Control (1.0 SMY)
- 1. White-fringed Beetles. At Gulfport, Miss., 35 chemical compounds were tested in a specially constructed glass olfactometer as attractants or repellents against adult white-fringed beetles. The beetles were not visibly attracted or repelled by any of the materials.

Grant research at the University of Georgia, Athens, Ga., showed that white-fringed beetle adults were not responsive to volatile components of intact or crushed leaves of favored host plants including peanuts, sovbeans, aster, cocklebur, or ragweed. Twenty-four chemicals were also tested but none showed any repellent or attractant activity.

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RPA 112 - RANGE MANAGEMENT

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RPA 207 - CONTROL OF INSECT PESTS OF FIELD CROPS

Basic Biology, Physiology, and Nutrition

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PRODUCTION AND HARVESTING EQUIPMENT AND METHODS Agricultural Engineering Research Division, ARS

USDA and Cooperative Program

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Corn	•		:				•		*				:	
Georgia	:				:	1.0	•		:				:	1.0
Iowa	:	2.0			:	0.3		0.7	:				:	3:0
Missouri	:		*		:		:	1.0	:		•		:	1.0
Ohio			:		:	0.4	:		:		:		•	0.4
Total Corn	:	2.0	:		:	1.7	:	1.7	:		:		:	5.4
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Small Grain	:		•		:		:		:		•		:	
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(Beltsville)	:		*		:		•		:		•	0.5	*	0.5
Georgia	:				:		:		:		:	0.3	:	0.3
Iowa					•		:		:		:	3.0	•	3.0
Nebraska					:	0.5	•		:		*			0.5
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Georgia	•				:		•					1.9	:	1.9
Oregon	:		:		:				:		:	4.0	:	4.0
Texas	:			1.0	:								:	1.0
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Problems and Objectives

Better methods, techniques, equipment and structures for use on the farm for producing, harvesting, storing, and the initial preparation of grain, seed and forage crops for market are needed to increase efficiency in the use of labor and equipment, preserve quality and prevent spoilage and damage from mechanical handling and insects. Electromagnetic energy has many established farm uses but research indicates other highly useful potential capabilities in farm production, such as attracting and killing insects and improving the germination of seeds. There is considerable present need for precise seedbed requirements for various crops in different areas of the country. The exact best placement for starter fertilizer is also unknown for a number of crops under different climatic and soil conditions. Every method to control or eradicate plant diseases, weeds, and insects requires some type of equipment. Effectiveness of the equipment necessary may be essential to the success of the method which is attempted or recommended. The cost of harvesting and farm handling of forage, grain and seed crops is the major expense of production. The supply and adequacy of manpower for these operations are becoming progressively less satisfactory, thus requiring additional research in this area. While considerable information has already been obtained for the development of processes such as drying and separation, basic and more precise information must be developed for these and other processes in order to achieve further progress. New trends in storage structures also require additional research in design and use.

The major objectives of research on production and harvesting equipment and methods for grain and forage crops are:

- 1. To determine the effect of soil mechanics and equipment mechanics on the tillage operation.
- 2. To develop tillage methods and systems of equipment which are compatible with conservation farming practices.
- 3. To develop methods and equipment for reducing brush cover on rangeland and for seeding grasses on rough rangeland.
- 4. To develop and evaluate better methods for applying insecticides.
- 5. To improve deposition efficiency of chemical sprays and reduce drift of spray particles.

- 6. To develop equipment and methods for treating plant materials with electromagnetic radiation characteristics of seed.
- 7. To develop machinery and improved machine components to plant, cultivate, harvest, and handle grain and forage crops in order to minimize power and labor requirements, reduce the number of field operations and provide optimum seedbed, growing and harvesting conditions.

Progress - USDA and Cooperative Program

RPA 102 - SOIL STRUCTURE AND SOIL, PLANT, WATER, NUTRIENT RELATIONSHIPS

A. Tillage Systems

Research at Ames, Iowa, in cooperation with the Iowa Agricultural Experiment Station, using 30-inch spacing of corn rows, indicated that applications Atrazine and Simazine were successful substitutes for plowing. Secondary tillage operations added little to corn yields when weeds were controlled and adequate stands were obtained. Strip-rotary tillage resulted in more consistent and uniform stands than other secondary tillage systems. These studies indicate that corn can be produced without tillage but some tillage is necessary to obtain satisfactory stands. Tillage system evaluations where weeds were controlled chemically show that stand establishment is the major problem. Results from 1964-1967 studies on tillage systems show that listing, no tillage, and till-planting result in fewer plants and less stand uniformity than the other systems. Evaluations of rotary and conventional tillage systems on the basis of inputs (time, fuel, labor and machinery costs) and outputs (corn yield) with restrictions on weather, acreage, and timeliness indicate a greater return per unit of investment with corn acreage above 500. Model studies of vibrating tools show some draft reduction but no reduction in total energy input. Soil clod size distribution was controlled by controlling amplitude and frequency of tillage tool vibration. Combinations of standard deviations and spectral density analysis of point elevation readings show promise as a method of describing soil surface roughness. Results from continuous corn and sovbeans grown on 30-inch ridges indicate the need for design changes in planter and fertilizer application equipment.

RPA 112 - RANGE MANAGEMENT

A. Grass Seeding and Brush Eradication

Five special machines designed and constructed by AERD were used to seed range grasses in cooperative field experiments at eleven different locations in North Central Texas. Four of six herbicides gave satisfactory

control of competitive plants in the establishment of a number of the ten species tested. Both broad leaf weeds and annual grasses were checked by atrazine, CP 50144, diuron and tritac (all at 1 1/2 lbs./A). Four species tolerated the first herbicide -- three species the next two chemicals. (Jose tall wheat grass and sand bluestem were in both groups.) In the range grass studies in the Canadian River Valley, only two or nine species were able to get satisfactory establishment (Ermelo lovegrass and blue panic). In the application of nitrogen fertilizer to native range grasses in the same area, production from treatments with 50 lbs. N/A subsurface were equal to 200 lbs. N/A broadcast.

Contract research with the New Mexico Agricultural Experiment Station on range seeding equipment was concluded and the project closed.

Five acre test plantings were seeded in June 1967 at five southwestern U.S. locations with equipment previously developed on the project. This tractor-mounted and drawn equipment uproots undesirable brush, seeds desirable range grasses, and repositions the uprooted brush on the seeded strip.

The equipment modification under test was a redesigned, overhead conveyor for handling the brush. Three additional plantings were made in July after further remodeling of the equipment.

The remodeled equipment functioned satisfactorily and accomplished the desired objective. However, it is heavy and awkward to maneuver. It will have to be redesigned to make it commercially acceptable, but such a combined conveyor-planter could have wide application in southwestern range land. The resulting stands of range grasses are to be evaluated later.

RPA 207 - CONTROL OF INSECTS

A. Corn

- 1. Georgia. Equipment to mechanize removal of fall armyworms from containers in the mass rearing operations was improved and effective. Oviposition cages were improved materially in reducing labor for the corn earworm pupae-rearing operations. Removal of small insect scale buildup in cages was not completely solved.
- 2. <u>Iowa</u>. In cooperative work at the Iowa Station, including ENT Division, first-brood borer control was not improved when rate of application of DDT or diazinon was increased to compensate for different row spacings and plant populations. These preliminary results indicate that available granular application equipment and techniques are adequate for acceptable borer control where narrow rows and high populations are used. Systemic insecticides for corn borer control continue to show promise. Chemicals placed 10" to side of the corn plant 2-4" deep with a mechanical

cultivation gave better control than applications at planting time with seed, 2" to side of seed, with starter fertilizer, or 2" to side and 2" below the seed. Diazinon granular applications caused slight to moderate amounts of phytotoxicity on 45 single-cross corn varieties and 20 inbred lines. This phytotoxicity had little or no effect on yield. Changes in application equipment and techniques for applying diazinon granules are not indicated. Both liquid and granular formulations of Bacillus thuringiensis gave excellent control of first-brood borers. Some liquid formulations tested would not stay in suspension and could not be applied as sprays. Some granular formulations were inadequately sized and could not be metered accurately. Either formulations must be improved or equipment must be redesigned for acceptable Bacillus applications. Covering diazinon granules with 1, 2, or 3" of soil at cultivating time did not improve corn rootworm control over surface applications. Similar studies in previous years have shown that some soil coverage is beneficial but depth is not important.

3. Ohio. At the Ohio Agricultural Research and Development Center in cooperative studies with ENT Division, a granular insecticide was incorporated to various depths by several types of tools. Best yields were obtained from surface incorporation by a scraper blade tool. Several granular insecticides were surface incorporated in a 6-inch width over the planter row by the scraper blade tool. Yield, root rating, and reduction of rootworm larvae population were significantly improved in Furdan-10g and Aldrin-20g treatments.

Liquid sprays of Baygon, carbaryl, diazinon, and malathion applied to the ear zone for control of adult northern corn rootworm each reduced population by at least 96 percent. Yields were significantly increased in each treatment compared to an untreated plot.

Corn leaf aphids were controlled by a spray of 57 percent malathion EC applied either over the row or from the sides of the row.

Laboratory study of granular applicators shows considerable linear variation in amounts of material distributed down the row even though total flow over a timed period may indicate the feed rate is uniform.

4. Corn insect response to infrared. In preparation for staffing the Insect Attractants Laboratory at Gainesville, Florida, a professional was hired and temporarily stationed at the Iowa Agricultural Experiment Station. There he pursued findings of others that the carbon dioxide (CO₂) production of an insect may be used as an indicator of response to different stimuli. Laboratory equipment and instruments have been assembled for measuring the CO₂ output of European corn borer adults while subjected to selected wavelength bands of radiation between 0.25 and 15 microns. Basic components are an infrared CO₂ analyzer and a monochromator. Air, free of CO₂, is passed through the insect chamber to the CO₂ analyzer and

exhausted. Preliminary experiments revealed patterns of behavior for single insects, showing suppression of activity by light and a maximum activity in darkness from 4 a.m. to 8 a.m. Suppression of activity by light varies with age of insect and time of day. During the dark nights, the characteristic night activity persists regardless of previous handling and exposure to light. Activity may last 10 minutes and resume after less than 1 minute of rest. CO2 output from inactive insects fluctuates very little; however, this output varies with temperature and other factors. Output of an active insect may be 20 times greater than that of an inactive insect. Outputs have ranged from 0.025 to 0.5 cc/hr/insect. A change of about 0.005 cc/hr. can be detected. The apparatus allows application of radiation to the insects in wide bands, using optical filters, or in narrow bands. Other stimuli, such as sound or a sex pheromone, can be introduced.

B. Stored Grain

Cooperative Studies of Radiofrequency Treatments with Nebraska AES. Radiofrequency (RF) electrical treatment of infested grain can control all developmental stages of all species of stored-grain insects tested thus far. Insect control can be achieved by treatments which do not damage wheat germination nor its milling and baking qualities, but costs of the electrical treatment are estimated to exceed those of chemical control methods. Therefore, studies are aimed at improving the efficiency of the RF control method. Work this year has dealt mainly with the development of methods for measuring the relative RF dielectric properties of insects and grain. These properties determine the relative absorption of energy from the RF electric field by insects and grain. Differences in the dielectric properties of the different materials determine the degree of selective absorption of energy by the insects as compared to grain. Since the dielectric properties of both vary with frequency, the measurement of the dielectric properties of insects and grain is necessary over a wide frequency range to discover the most effective frequencies to employ for insect control treatment of infested grain. Substantial progress was made this year in the development and improvement of methods for measuring dielectric properties in the following frequency ranges: 250 Hz to 20 KHz, 1 to 250 MHz, and 200 to 500 MHz. Some measurements were taken on adult rice weevils and on wheat samples, but analysis of results must

RPA 209 - CONTROL OF WEEDS AND OTHER HAZARDS

await completion of methods development for reliable interpretation.

A. Control of Weeds in Corn

1. <u>Soil incorporation of herbicides</u> for weed control in corn showed little benefit in studies at Ames, Iowa. Preplant applications were more effective than postplant. All incorporating tools were equally effective.

Early weed control was better when Treflan was incorporated. This improved weed control was not reflected in increased yields. Harrowing after planting and shallow cultivations were not needed when timely cultivations with sweeps and disk hillers were used to control weeds in corn. The germination rate of seeds tended to be decreased when subjected to high intensity electrical fields. Analysis of soil and water samples from controlled runoff plots showed some loss of pesticides in both soil and water. Nontoxic forms of Ramrod and Diazinon were found within two weeks after application. Directional and overall applications of Atrazine and oil were equally effective and both gave excellent weed control in corn when followed by mechanical cultivations. Weeds were successfully controlled with combinations using multiple row spacings for corn where a roadway was left for tractor tires.

Effect of directional spray equipment. At Columbia, Missouri, directional spray equipment had no effect on weed control in corn with Lorox and Clobber. Leaf lifters reduced damage when corn was less than 12 in, high. Directional and overall applications of Atrazine and oil gave equally effective weed control in corn rows. Lack of control between rows with directional sprays caused yield reductions. Directional spray studies with DNBP show that application equipment had little or no effect on control; however, soybeans less than 6-8 inches tall were damaged. Surface applications of Sutan-D were equal to incorporation with a powered rotary cultivator or injected beneath a sweep, but corn damage occurred with sweep applications. The effectiveness of Amiben was not improved with incorporation. Preemergence herbicides showed improved weed control when followed by rotary hoeing at spike stage. Fan-type nozzles gave better weed control than air atomizing nozzles for 0.25 to 1.0 gpa applications of Amiben and trifluralin. Variations in trifluralin between individual granules was 3 percent with a random selection from a 5 percent product.

RPA 307 - IMPROVEMENT OF BIOLOGICAL EFFICIENCY

A. Seed Treatment

1. Radiofrequency treatment. Work in cooperation with the Nebraska Agricultural Experiment Station at Lincoln confirmed the effectiveness of radiofrequency (RF) electrical treatments for safely reducing hard-seed content in alfalfa. Tests with 27 different seed lots showed that RF and infrared treatments were equally effective for reducing hard-seed content. Furthermore, RF-, infrared-, and glow-discharge-treated seed exposed to these three different types of electrical treatment 5 years ago germinated well in retesting, and beneficial effects of the three types of treatment were still evident. Seed quality had not deteriorated in storage as it does with mechanically scarified alfalfa seed.

Studies initiated this year on use of RF energy for drying of chopped alfalfa revealed that RF treatment for exposures longer than 30 seconds measurably accelerated the subsequent drying rate of alfalfa. No changes in protein content of the dried chopped alfalfa were found, but the carotene retention of the dried alfalfa was doubled by some RF treatments. Experimental RF treatment of pine seed revealed a noticeable acceleration of germination for seed lots of Digger and loblolly, but further work is necessary to properly evaluate the benefits of treatment. In tests with seeds of desert plants, germination of huisache was raised from 6 percent to 30 percent by RF treatment of samples conditioned at 0 degrees F. before exposure to RF electric fields. Substantial increases in germination of castorbeans were obtained with RF seed treatment.

RPA 308 - MECHANIZATION OF PRODUCTION OF FIELD CROPS

A. Forage

Planting equipment. In cooperation with the Georgia Agricultural Experiment Station, four specially developed machines were used to seed seven cooperative field experiments of forages at three locations in the Southeast. Some were on prepared seedbeds while others were interseeded in sods. In selecting suitable cool growing forages for Midland bermudagrass sods, eight permanent type species were interseeded. In the row spacing studies of tall fescues interseeded in hybrid bermudagrass in mountain areas (Midland), the cool growing grass produced more dry matter than the summer grasses. In the 8-inch spacing, the dry matter production was over 5 tons/acre, with the fescue outproducing the bermudagrass from four to six times on the monthly clippings. In the 16-inch row spacing, the forage production of fescue averaged two to three times that of bermudagrass. With 24-inch row spacing, bermudagrass slightly out-produced the fescue. All treatments produced approximately the same total tonnage regardless of the row spacing of the interseeded crop. One substantial advantage of the dual crop is in the saving of labor in livestock feeding by extending the pasturing time from three to six months.

At Beltsville, Maryland, three field experiments were conducted with a special sod interseeding machine. Some twelve different annual forages were interseeded in cool season grass sod (tall fescue). In the interseeding of winter grains in Midland bermudagrass, the dry matter production increased about 80 percent (2.9 tons/acre to 5.2). The total production increase was not the only increased benefit. Having forage during a longer period of time (in late fall and early spring) is a marked advantage to a farm enterprise as it reduces materially the amount of labor required in feeding livestock. A report was analyzed and completed in 1967 of an earlier series of field experiments to determine the limits of successfully establishing spring seeded grasses and legumes. Although the primary objective was not achieved for the two seasons studied, a definite correlation was attained between soil moisture conditions and survival of

herbage stand. A formula was derived whereby the survival stand could be predicted from a given soil moisture condition during the period following the seeding.

- 2. Harvesting and farm processing Coastal bermudagrass. For the second consecutive year, there was no significant difference in either yield or protein content between irrigated and nonirrigated Coastal bermudagrass. Near isogenic lines of dwarf and tall millet were dehydrated. Although the short millet had a much higher ratic of leaves to stem, the stem was more succulent than that of the tall millet and there was no difference in processing energy requirements. Continued studies of most economical method of producing pellets for roughage indicate that small amounts of molasses and animal fat can be economically used.
- 3. Seed harvesting equipment and methods. An experimental double belt thresher was designed, constructed, and tested. Variables of belt texture, speed, clearance, and pressure, as well as rate of feed, were investigated. Tests with two legume and three grass crops on a one-foot wide belt unit gave capacities up to 1600 lb./hr. at the 90 percent threshed seed level. Seed damage was judged negligible. A thresher with a four-foot wide belt will have capacities comparable to a six-foot swath spike-tooth cylinder combine. Time-of-harvest studies in 1967 indicated that the optimum mowing time for creeping red fescue was at a seed moisture of 23 percent; for Chewings fescue, 40 percent; and for perennial ryegrass, 40 percent.
- Seed processing equipment and methods. Crop seeds and contaminants from 18 seed mixtures were measured microscopically to help select optimum size screens or indents for removal of contaminants. Full-size units of the resilience separator and vibrator separator were constructed and are operating in processing plants. Magnetic cleaning studies showed that water levels were critical in removing contaminants from alfalfa and directly related to clean seed yields. Particle size distributions and performances of 10 iron powders showed excellent agreement between fineness and effectiveness of a powder in removing contaminants. Flat-shaped particles did a better job than blocky shapes of the same general size. Reuse of powder was found inefficient because fine particles were gradually removed, leaving progressively coarser powder. Investigation of seed feeders was continued. The first phase of these studies was observation of oscilloscope waveforms showing accelerations of the various vibrators. The scope signal was obtained from a piezoelectric accelerometer mounted on the vibrator tray. Some of the traces were photographed with a specially constructed Polaroid camera setup. A uniform flow seed feeder was developed. Direct drive by a synchronous motor insured a stable frequency for the vibrator tray and tests showed very uniform feed rates, even with changing temperature or voltage. A high-speed seed counter was developed making use of a photocell sensing circuit and an electronic counter.

B. Grain

- 1. Fertilization. In continuing the study of row spacing and fertilizer placement with small grains, some of the new high producing varieties were used this year. In this first year study, with NC 4672 (Blueboy) wheat variety, 4-inch row spacing contact fertilizer placement increased yields about 1/3 (31.5 bu./A to 42.0) over the common practice of 8-inch row spacing (fertilizer in contact with seed). But of the treatments with side placement of fertilizer, the 12-inch row spacing had the highest yields (38.5), which also was a greater yield than the 8-inch or 12-inch row spacing with fertilizer in contact with the seed. In comparing fertilizer application positions on spring wheat (SWC-Montana) the 8-inch depth, continuous band gave highest yields compared to 4-inch depth and surface applications. Starter fertilizer with seed (4 lbs. N and 4P) in general depressed production.
- 2. Drying principles and methods. Final analysis of data on counterflow drying was made and a report prepared. Tests showed drying time is related to initial and final corn moisture and to air temperature and humidity. Criteria were presented for optimizing drying capacity and thermal efficiency. No damage to germination was observed using air at 140° F. compared to maximum of 110° F. for conventional drying. Weekly samples of corn obtained from field were taken to show changes in moisture as corn matures. Samples were processed to study effects of heated air drying on dry matter content, test weight, and germination.
- Quality deterioration during harvest, drying, and storage. A machine to shell corn by squeezing and rolling action was developed and tested. Ears were fed between two 12-inch wide opposing flat rubber belts traveling in opposite directions and at different speeds. Complete shelling was achieved at 35 percent kernel moisture. Shelling corn of 25 percent moisture resulted in less than two percent mechanical damage by visual inspection compared to 30 percent damage by current field harvesting machines. Mechanical damage is defined as any crack or minute fracture in seedcoat. Several sources of variation affect precision of visual inspection for determining mechanical damage in corn, including sample differences, differences among men, and man's inability to repeat damage estimate. Fast green dye did not improve precision. Studied relation of corn deterioration (measured by carbon dioxide production) to aflatoxin development. Of 72 samples, 10 had Aspergillus flavus mold, all 10 from corn above 20 percent moisture. Only two samples had aflatoxin after producing 22 and 28 grams of carbon dioxide per kilogram of dry matter and were very moldy in appearance. Initiated test of refrigerated storage of high-moisture shelled corn. A 6000-bushel bin was filled with corn at 21 to 23 percent moisture in November and cooled to 35° F. within three days; to below 10° F by January; and was still 27° F. on April 1. Initial test of two experimental chemical fungicides with 32 percent moisture shelled corn in storage indicated retarded mold development for one fungicide.

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PROGRESS REPORT

on

GRAIN AND FORAGE CROPS RESEARCH

of the

UNITED STATES DEPARTMENT OF AGRICULTURE

This progress report includes a summary of the current research of the USDA on grain and forage crops and a preliminary report of progress made during the preceding year. It is primarily a tool for use of scientists and administrators in program coordination, development and evaluation; and for use of advisory committees in program review and development of recommendations for future research programs.

The summaries of progress on USDA and cooperative research include some tentative results that have not been tested sufficiently to justify general release. Such findings, when adequately confirmed, will be released promptly through established channels. Because of this, the report is not intended for publication and should not be referred to in literature citations. Copies are distributed only to members of Department staff, advisory committee members and others having a special interest in the development of public agricultural research programs.

This report also includes a list of publications reporting results of USDA and cooperative research issued between July 1, 1967, and June 30, 1968. Current agricultural research findings are also published in the monthly USDA publications, Agricultural Research and The Farm Index.

RESEARCH PROGRAM DEVELOPMENT AND EVALUATION STAFF
UNITED STATES DEPARTMENT OF AGRICULTURE

Washington, D.C. December 31, 1968

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II. NUTRITION, CONSUMER AND INDUSTRIAL USE RESEARCH

CORN UTILIZATION

Northern Utilization Research and Development Division, ARS

USDA and Cooperative Programs

Location and Nature of		: Scientist Man-Years FY 1968: Research Problem Area :			Total
Intramural Work	406		702 :		1000.1
Peoria, Illinois	•		•	a •	
Food A. Chemical Composition and Physical Properties B. Color, Texture and Other Quality Factors C. Microbiology and Toxicology D. Technology—Process and Product Development	9.7 2.5 0.9		6.5		9.7 2.5 7.4
Industrial Products	•	:	•		
A. Chemical Composition, Physical Properties and Structure B. Chemical and Physical Investi-		9.0	:	•	9.0
B. Chemical and Physical Investi- gations to Improve Products C. Microbiology and Fermentation	•	12.0	:	1.8	13.8 9.2
D. TechnologyProcess and Product Development	•	5.6	:	•	5.6
Total	:14.9	: 35.8 :	6.5 :	1.8 :	59.0

Intramural program is supplemented by extramural support representing (a) 0.7 SMY's at State Agricultural Experiment Stations, (b) 13.4 SMY's at other U.S. institutions, and (c) P.L. 480 funds in 10 countries representing 975,423 U.S. dollars equivalent.

- 1/ RPA 407: 0.3 (Chemical and Physical Investigations to Improve Products)
 RPA 702: 0.4 (Microbiology and Toxicology)
- 2/ RPA 406: 3.6 (Chemical Composition and Physical Properties, 1.3; Microbiology and Toxicology, 2.3)
 - RPA 407: 8.7 (Chemical Composition, Physical Properties and Structure, 1.2; Chemical and Physical Investigations to Improve Products, 2.7; Microbiology and Fermentation, 1.5; Technology--Process and Product Development, 3.3)
 - RPA 702: 0.2 (Microbiology and Toxicology)
 - RPA 901: 0.9 (Chemical and Physical Investigations to Improve Products)

Problems and Objectives

The farmer sells 100,000,000 bu. of grain to make 3 billion pounds of cereal starches and flours that are used annually in the U.S. for industrial purposes. Corn is the source of most of these products. These outlets are constantly threatened by synthetic products derived from nonagricultural sources. Increasing the present and future competitive position of corn starch and flour in industrial markets requires development of new and improved products that will do a better job in outlets such as the paper industry, industrial chemicals, adhesives, protective coatings, plastics, elastomers, and thickening agents. Almost 80 percent of the U.S. annual production of corn is used as animal feed. Because of the extremely large volume of this outlet, even small improvements in quality or processing efficiency give direct gains to industry and to the farmer. Utilization of corn in foods also is an outlet of great economic importance. Significant benefits can be realized if we can make cheaper and more nutritious products, including foods from new corn varieties having higher nutritional value than does ordinary dent corn.

Major objectives of the current research are to develop and evaluate alternate ways to:

- 1. Discover new technology for converting starch by chemical or fermentative means to products for use in the paper, chemical, and other industries.
- 2. Provide increased industrial outlets for new classes of high-amylose corn.
- 3. Improve yields and quality of corn dry milling fractions.
- 4. Lower the cost of converting corn to sugars and sirups and to more acceptable, varied, and nutritious foods.
- 5. Remove mold toxins or harmful microorganisms from corn and corn products.

Progress - USDA and Cooperative Programs

RPA 406 - NEW AND IMPROVED FOOD PRODUCTS FROM FIELD CROPS

A. Chemical Composition and Physical Properties

1. <u>Corn proteins</u>. Gel filtration chromatography on Sephadex G-100 at a pH of 8.6 has yielded several fractions from germ and endosperm albumins. Extensive aggregation at this pH was evidenced by the elution volume of the bulk of the proteins in contrast to results at pH 4.4. Subcellular particles from immature corn endosperm homogenates in phosphate buffer were

separated by ultracentrifugation on a sucrose density gradient. The major protein of the protein bodies was identified as zein. Removal of the zein with 70 percent ethanol extraction resulted in a residual protein with unique amino acid and electrophoretic properties after reduction. This protein resembles certain of the glutelin components isolated by direct extraction from endosperm. Amino acid and protein analyses of highlysine corn kernel fractions showed that germ cake, bran, and aspiration meal have high protein content and good amino acid balance. The aleurone layer, comprising 2 percent of the grain, contained 30 percent protein, predominantly albumins and globulins rich in lysine.

Various methods of tryptophan analysis have been evaluated. S(pyridylethyl)-L-cysteine has been prepared and characterized for use as an internal standard in amino acid analysis. A computer program, using linear programming, has been written which gives the best nutritionally optimum combination of cereal grains based upon their essential amino acid patterns and cost.

Grant research at Purdue University, Lafayette, Indiana, showed that water-soluble proteins of corn are separable by chromatography on DEAE-cellulose columns. An improved method for tryptophan analysis that provides accurate reproducible values has been developed which will allow routine analysis for this amino acid in corn. A larger amount of dialyzable nitrogen extracted with saline solution from high-lysine corn than from normal corn was found to be due to a high level of nonprotein nitrogen. Most of this nonprotein nitrogen was attributable to free amino acids.

- 2. Corn lipids. Two hybrid yellow corns were hand-dissected into pericarp (4% of the whole kernel), tip cap (4%), floury endosperm (22%), horny endosperm (63%), and germ (7%). Hexane-soluble material in the whole corn averaged 3.6 percent. Thin-layer chromatograms of nonpolar lipids and the hand-dissected fractions were scanned by a densitometer. Average values for the various lipid fractions as percent of the total lipids in the hexane extract were: phospholipids, 1; diglycerides, 3; sterols, 3; unknown "A", 2; fatty acids, 6; unknown "B", 5; triglycerides, 73; and hydrocarbons and sterol esters, 7. In the hand-dissected fractions, unknown "A", diglycerides and sterols made up 1 to 6 percent of the lipids. Fatty acids ranged from 2 to 14 percent and unknown "B" from 0.5 to 12 percent. Triglycerides, the major fraction, accounted for 55 to 88 percent of the total lipids. Hydrocarbons and sterol esters were five times more concentrated in the horny endosperm than in the floury endosperm.
- 3. Reactions of maltose and glucose. Intermolecular hydrogen-bonded hydroxyl groups, as shown by X-ray structural studies of maltose, lactose, and cellobiose, persist in lowly polar solvents and now appear to be detectable by chemical methods. Heterogeneous acetylation of α or β -maltose gave a 70-percent yield of 1,2,6,2',3',4',6'-hepta-0-acetyl maltose with only a 25-percent yield of the octaacetate. The corresponding heptaacetates of α and β -lactose and of β -cellobiose were formed in 50-percent yield. Isomaltose and melibiose (not intramolecularly hydrogen-bonded) were almost completely acetylated under identical conditions. The α -anomer of

isomaltose, previously unknown, complexes in crystalline form with either sodium iodide or sodium bromide. It was recovered in 92-percent anomeric purity. Large β-maltose hydrate crystals suitable for X-ray structural studies were obtained in 99-percent anomeric purity. Phase diagrams of the melting point of intimate mixtures of anhydrous maltose or D-glucose with urea show that maltose forms 4:1, 2:1, and 1:1 molecular complexes, whereas D-glucose forms only a 1:1 complex. A 1:1 complex of maltose with imidazole was precipitated from ethyl acetate solution, but D-glucose did not give an isolable complex under similar conditions.

- 4. Structural and chemical differences among corn genotypes. Alcohol solubility data and microscopic analysis have shown that opaque-2 and floury-2 high-lysine maize mutants were deficient in granular protein, the main site of zein deposition in maize endosperm. Subcellular bodies similar to zein bodies of normal maize are lacking in the protein structure of floury-2 mutant. In opaque-2, small protein bodies averaging about 0.1 µ in diameter were noted 14 days after pollination. The size of these bodies was unchanged in samples collected at 20 and 50 days. In contrast, zein bodies of normal corn showed a 10- to 20-fold increase in size over this period. The two mutants responded differently to gene dosage. Endosperm subcellular structure in the floury-2 mutant developed subcellular protein structure similar to the homozygous condition with one dose of the floury-2 gene. In opaque-2, well-developed zein granules persisted in endosperm protein with one and two doses of opaque-2 gene.
- 5. Phytin in corn. Based on studies of wet- and dry-milled corn products obtained from industrial organizations, preliminary results from grant research at the University of Missouri, Columbia, Missouri, indicate that over one-half of the phytate in corn is present in the germ.

B. Color, Texture and Other Quality Factors

1. Browning reactions. Dry heating of the Amadori compound, 1-deoxy-1piperidino-D-fructose (a model for cereal browning intermediates), under vacuum at $1\overline{10}^{\circ}$ C. gave a sublimate that contained five major and at least eight minor components. Nine compounds were cleanly separated by column and gas chromatography, and their structures were determined by nuclear magnetic resonance, infrared and mass spectrometry, and synthesis. Major components were piperidine acetate, piperidides of carbonic, formic, and acetic acids, piperidino-C-methyl-triose-reductone (enolic 1-piperidinobutanedione-2,3), and α -piperidino- Υ -butyrolactone. Minor components were piperidides of glycolic and lactic acids, and an isomeric piperidino-Cmethyl-triose reductone. A trace fraction with fragrant caramel aroma was separated into nine components, several of which have been tentatively identified. The amino hexose chain-fission gave predominantly 2- and 4-carbon fragments, and not the 3-carbon fragments that predominate from unsubstituted hexoses in aqueous alkaline solutions. Origin of the most powerfully fragrant caramel aroma compound, 4-hydroxy-2,5-dimethy1-3(2H)furanone, now detected in several foods, was traced to a 1-amino-1-deoxy-2ketose (Amadori compound) intermediate.

C. Microbiology and Toxicology

1. Studies on enzymes. In studies on enzymes under a grant to the University of Nebraska, Lincoln, Nebraska, two Aspergillus niger glucoamylases (I, II) showed molecular weights of 125,000 by sedimentation velocity techniques while sedimentation equilibrium analyses indicated 80,600 for I and 58,500 for II. Significantly, this value for II was only about one-half the value of 98,000 obtained by N-terminal amino acid analyses. Carbohydrate moieties of both isozymes appear to be linked through D-mannose to serine or threonine and to be present as small oligomeric units.

Under a grant to the University of Arkansas, Fayetteville, Arkansas, studies on labeled oligosaccharides have permitted the assignment of relative binding affinities to the various subsites on <u>Bacillus</u> subtilis amylase. The active site spans about nine glucose residues and appears relatively rigid. The rate of cleavage of a given glycosidic linkage is presumably independent of the size of the substrate. Attempts to label the active site of lysozyme with radioactive cellohexose appear to be successful.

At Iowa State University, Ames, Iowa, also under grant, a strain of Streptococcus equinus was found which produces an extracellular amylase but is not repressed by glucose as is extracellular amylase production in most Streptococci. Strains of Pseudomonas stutzeri produced maltotetraose almost exclusively from amylose and amylopectin. Ps. saccharophila produces maltose and maltotriose as well as lesser amounts of higher dextrins. Numerous strains of Bacillus have been screened for amylolytic properties. Some appear to resemble pancreatic α -amylase while others yield typical B. subtilis-type α -amylase. Some strains of B. polymyxa produce predominantly maltose as the end products. One polymyxa strain gives oligosaccharides of three and five glucose units in length.

In research on the purification of enzymes, under a grant to Kansas State University, Manhattan, Kansas, Sephadex G-200 Superfine was found to be a satisfactory anti-convection agent for preparative disc electrophoresis. Chromatographic separations of mixed proteins in near gram quantities are now possible. A new column design permits stacking phase separations in which the resolved sample may be removed by liquid flow into fraction collector tubes. A system of electrolyte materials has been developed which employs an intrinsically slow species of unit charge and extends the separating abilities of the system to a wider variety of proteins. The system operates with lower current requirements, thus preventing heating of the columns.

2. Taxonomic and related investigations. Zygospores were discovered for the first time for <u>Rhizopus oryzae</u>. NRRL 3133, which is probably Went's type strain for the species, formed up to 277 zygospores when mated with three other strains of <u>R</u>. <u>oryzae</u> in the collection. As many as 550 zygospores were formed when NRRL 3133 was mated with a strongly mating strain

of R. microsporus, which in turn gave over 1,000 zygospores per platemating with NRRL 2915, the type strain of R. microsporus.

Under a grant to the University of Minnesota, Minneapolis, Minnesota, initial work has been primarily concerned with the development of methods. Photographic procedures for studying hyphal growth and methods for determining enzymatic activity of hyphal sections are being established.

D. Technology--Process and Product Development

- 1. Corn dry milling. Cold tempering and degermination tests on a second lot of high-moisture corn (19%) confirmed previous results indicating a minimum change in degerminator response with change in temper time. Few stress cracks were found in air-dried -4+5 grits prepared from 21-percent-moisture corn under a variety of tempering conditions, a small number from 17-percent corn, and an appreciable number from 13-percent corn. An exploratory, two-step, simulated, series-type operation of tempered corn through a Beall degerminator increased yield of -4+6 grits by 25 percent over a conventional one-pass operation, but did not decrease production of undesirable fines. Treatment of whole untempered corn in a 5-percent caustic solution for 5 minutes followed by passage through a modified sanitary pump successfully removed much of the hull. After tempering and degerminating, the dehulled corn produced 5 percent less fines, 10 percent more grits, and 0.1 to 0.4 percentage point less oil in the grits.
- 2. Mechanical and viscoelastic properties of corn kernels. In studies at Pennsylvania State University, University Park, Pennsylvania, a slab of horny endosperm (12% moisture, w.b.) from a corn kernel was found to have a complex frequency-dependent modulus (E) of 12,800 pounds per square inch. E, evaluated at 1 cycle/second, was found to decrease with increasing moisture content over the range of 7 to 25 percent (d.b.) with a large drop occurring at the 16-17 percent moisture content level. The expansion coefficient for horny endosperm has been experimentally determined to be approximately three times as great for a 1 percent change in moisture content as for a 1° F. change in temperature. A complex equation has been developed for predicting conditions leading to the formation of undesirable stress cracks during the process for artificially drying corn.

RPA 407 - NEW AND IMPROVED FEED, TEXTILE AND INDUSTRIAL PRODUCTS FROM FIELD CROPS

Industrial Products

- A. Chemical Composition, Physical Properties and Structure
- 1. Physical and chemical studies on starch components. Detailed studies on the optical rotation of starch components in the solvent system H₂O-DMSO, needed for the determination of concentrations in solution, are nearing completion. Experimental studies show a discontinuity in the specific rotation corrected for refractive index variation at a solvent

composition corresponding to the proposed complex $2H_2O$ -DMSO. This discontinuity is exhibited by α -1,4-linked polysaccharides such as corn and potato amylose and amylopectin. Optical rotation studies are underway on model sugars, and hydrodynamic studies on amylose are being made to determine whether the discontinuity corresponds to a polymer conformation change or a solvent effect.

The oriented structure " $V_{\rm DMSO}$ " obtained from dimethyl-sulfoxide-cast amylose films may be transformed to other "V" structures by appropriate chemical treatment. Unit cells and the probable space group of $P2_12_12_1$ were determined for the "V" anhydrous and "V" hydrate forms of amylose. The analysis confirmed previous predictions by Rundle and coworkers. Complexes of amylose with <u>n</u>-butanol were prepared and subjected to X-ray analysis. Their unit cells were calculated and the space group $P2_12_12_1$ was confirmed. No evidence exists for methanol, ethanol, or <u>n</u>-propanol structures similar to that shown by the <u>n</u>-butanol complex.

An NMR method for determining the position of S-benzyl xanthate in a series of α -D-glucopyranoside derivatives has been developed and successfully used to differentiate the 2-, 3-, and 6-substituted isomers of a prepared series. The technique should be applicable to the study of other electronegative substituents. Deuterium resonance has been applied successfully to the problem of determining the deuteration site in experiments carried out under the same conditions as hydrogenation.

Scattering patterns from single starch granules are being investigated under a grant to the State University of New York, Syracuse, New York. Higher order maxima predicted by theory have been observed for canna starch granules dispersed in an imbibition liquid of matching refractive index. Work has begun on preparation of amylose films with both oriented and unoriented specific crystalline structure. An oriented "B" X-ray pattern amylose film is being used to determine the sign of birefringence for the oriented "B" structure.

In studies under a grant to Arizona State University, Tempe, Arizona, it was found that the reaction between "V" amylose hydrate and HCl yielded more constant ratios of products than did the reaction between amorphous amylose and HCl. This result indicates that the sites offered in the crystalline form are more selective in reactions with small molecules. A Patterson function of KOH-amylose is being constructed. As a main feature it has a large peak at (1/2, 0, 1/2) which is interpreted to mean that only a limited number of helix orientations are possible in the $P2_12_12_1$ space group.

Research conducted under a PL 480 grant to the University of Osaka Prefecture, Sakai, Japan, showed that the reaction of o-phenylenediamine with glucose, maltose, and related oligosaccharides gave quinoxalines that can be determined polarographically. Reaction conditions are being sought for applying this reaction to the determination of carbonyl groups in starch and its derivatives.

Research under a PL 480 grant at the University of London, London, England, showed that pullulanase, the extracellular glycogen and amylopectin debranching enzyme of <u>Aerobacter aerogenes</u> is unusually resistant to heat inactivation. Its activity is totally destroyed by heat above 70° C. However, a considerable portion of this activity can be regained on storage at room temperature. The extent of reactivation depends on pH, enzyme concentration, and the specific enzyme preparation. The last variable appears to be the result of at least two factors. First, pullulanase preparations contain both a high and low molecular weight species. The presence of the former is necessary for the stabilization of the latter. Second, the preparations also contain a dialyzable glycoprotein (M = 6,000-10,000) which is necessary for stabilization of the enzyme. The proportion of these species in different preparations is variable. This research has been completed.

2. Structure and properties of polysaccharides. A highly sensitive microtechnique has revealed further evidence of the composition of polysaccharide Y-6272. Analysis of a length-of-hydrolysis series for rate of appearance of intermediate and final products revealed (1) N-acetyl-glucosamine as the initial product with rapid conversion to glucosamine, and (2) N-acetylhexosaminuronic acid and a disaccharide as intermediates to final products which appear to be hexosaminuronic acid and glucosamine. Substituent groups in polysaccharides from Arthrobacter strains B-1797 and B-3225 have been identified as: pyruvic acid 5.5%, and 0-acetyl 7.8% for B-1797; and pyruvic acid 5.1%, O-acetyl 5-6% (tentative), and O-succinyl 2-3% (tentative) for B-3225. Reinvestigation of Y-1842 phosphomannan structure uncovered two new components: a lightly phosphorylated polymannoside (mannose/P = 29) and a tetrasaccharide monophosphate. The α -mannan component (mannose/P = 29; α_D + 61°) appears to consist largely of 1,2-linked units. An improved procedure was devised for isolation of the mannosyl glucose end group from Y-2023 phosphomannan. The Y-2023 polymer was found to contain 2-0- $(\alpha$ -Dmannosyl)-D-glucose, a new disaccharide.

Investigations showed that polysaccharide Y-2448 behaves in 1 $\underline{\text{M}}$ KCl like a high-molecular-weight polyelectrolyte ($\overline{\text{M}}_{\text{W}}$ 20 X 10°) whose conformation closely resembles that of a random coil. Viscosity studies of polysaccharide B-1459 in 4 $\underline{\text{M}}$ urea and 0.01 $\underline{\text{M}}$ ammonium acetate indicate high intrinsic viscosity. From incomplete sedimentation-diffusion studies, the molecular weight presently is estimated to be between 6 X 10⁵ and 2 X 10°.

At the University of Wisconsin, Madison, Wisconsin, grant research showed that the degree of carbanilation of polysaccharides, as measured by incorporation of phenyl groups, could be readily determined by the use of $^{14}\text{C-labeled}$ phenyl isocyanate. Standard deviation is about 1 percent. The carbanilylated polysaccharide was completely deacetylated and depolymerized by treatment with 1.3 $\underline{\text{N}}$ HCl in dry 2-methoxyethanol at 56-59° for 6.5 hours. Methylation of the free acid form of polysaccharide B-1973 proceeded to 95 percent of theoretical in one step.

Under a PL 480 grant to the National Sugar Institute, Kanpur, India, scaled-up, preparative isolation (1 to 3 gram amounts) of several aldobiuronic acids and of an aldotriuronic acid has been achieved. Additional materials made ready for further use are a purified polysaccharide and the mixed barium salts of two additional polysaccharides.

B. Chemical and Physical Investigations to Improve Products

1. Physical properties of starch dispersions. At the University of Graz, Graz, Austria, most of the measuring techniques and theoretical basis required for obtaining valid rheological data on concentrated (order of 5 percent) starch-based dispersions has been established. "Diagrams of state" have been prepared that graphically relate viscosity to both time dependent effects (thixotropy) and shear rate dependence. Dilute solution viscometry has been applied to characterize the starch materials in usual, absolute terms. Other properties being determined on pastes are gel strength and shear modulus (elasticity). Shear rates encountered in commercial uses of starch in coating formulations have been determined and will be used in experimental work. This study is being conducted under a PL 480 grant.

In this phase of work under a PL 480 grant at the Slovenian Academy of Sciences and Arts, Ljubljana, Yugoslavia, corn, wheat, and sorghum starches of American origin were modified by a variety of physical and chemical treatments designed to effect changes in their physical properties without destroying the granule structure in order to impart to the starches new and useful paste properties. Treatment of American starches with moisture contents of 10-23 percent by heating at 100°-120° C. for 3-5 hours or at 55° C. for 48 hours with 0.1 N mineral acids or sodium phosphate generally produced the greatest changes in their gelatinization temperature, paste viscosity, setback, solubility, swelling power, and transparency. However, for corn starch, modification both with dilute acid or sodium phosphate and with ultrasonics (1,600 KHZ; 3w/cm²) for 2 minutes was needed to produce the greatest change in paste solubility, swelling power, and transparency. Some detectable differences were found between X-ray patterns of the unmodified starches and those modified with acid or heat. All modifications were carried out without significant change in granule structure of the starch.

2. New derivatives of starch and related carbohydrates. The crystalline solid recovered from the reaction of bromoacetaldehyde diethyl acetal with D-mannitol was determined to be a 2:1 mixture of cis-1,2:cis-5,6-di-O-(bromoethylidene)-D-mannitol and cis-1,2:trans-5,6-di-O-(bromoethylidene)-D-mannitol. Studies on 1,2-O-bromoethylidene-D-mannitol established that the mono bromoethylidene acetal formation was equilibrium controlled and that conversion to the dibromoethylidene acetal was kinetically controlled and irreversible. Acetals formed irreversibly are determined by the stereo-chemistry of the transition state and relative rate of formation. As a result, they may not conform to the Hahn-Hudson rules.

Studies showed that the cyclic carbonate group formed from vicinal <u>trans</u> hydroxyl groups in pyranoid rings readily undergoes ring opening with various simple nucleophiles. The <u>trans</u>-fused cyclic carbonate and thiocarbonate groups also react with the hydroxyl groups of starch under various conditions to yield a starch product with pendent carbohydrate residues. An easy route was established for covalently bonding amino acids to carbohydrates, and conditions were found for the polymerization of carbohydrate <u>trans</u>-carbonates.

Grant research at the Ohio State Research Foundation, Columbus, Ohio, showed that reaction of dialdehyde starch and nitromethane yielded the expected product formed by addition of nitromethane to the aldehyde groups. Reduction of the nitro groups in this product to amino groups has not yet been achieved. Conditions were defined for the reaction of amylose with sulfuryl chloride to obtain a product completely chlorinated at C-6 and containing a cyclic sulfate. This product was subjected to hydrazinolysis, reduction with Raney nickel, and dialysis to yield an aminated amylose of D.S. 1 which had a low sulfur content (0.2%).

A synthetic method for preparing p-di(epoxyethyl)benzene has been developed under a grant to the University of Arizona, Tucson, Arizona. It involves reduction of p-di(chloroacetyl)benzene to p-di(α-hydroxyl-β-chloroethyl)-benzene with subsequent treatment with ethanolic potassium hydroxide. Studies show that diepoxides and polyols react to yield polymers which are probably crosslinked heteropolymers. Both m- and p-di(epoxyethyl)benzene have been synthesized to study effects of positional isomerism on the diepoxide-polyol polymerization. Synthesis of the ortho isomer proved to be impractical. This work is part of a study of the formation of polymers by reaction of diepoxides with starch and related carbohydrates.

Under grant research at Purdue University, Lafayette, Indiana, phosphine and phenylphosphine were added to the terminal olefinic bond of 5,6-dideoxy-1,2-Q-isopropylidene- α -D-xylo-hex-5-enofuranose under ultraviolet radiation. Two convenient routes for introducing nitrogen at carbon five of D-glucose, beginning with 5-Q-tosyl derivatives of 1,2-isopropylidene- α -D-glucofuranose, were found. These reactions have led to D-glucose derivatives with phosphorus or nitrogen replacing the pyranose ring oxygen, thus providing properties totally different from those of normal sugar.

In studies under a grant to the Ohio State Research Foundation, Columbus, Ohio, acetylated 1-thioaldopyranoses having the D-gluco, D-galacto, D-xylo, D-ribo, and D- and L-arabino configurations were synthesized and characterized for work on glycosulfenyl halides which are derived by halogenation of the thio sugar derivatives. New cyclopropyl derivatives, prepared from unsaturated sugars, were characterized. An easy method for preparing dimethylthiocarbamates, useful in simplifying NMR spectra, was demonstrated. These derivatives also offer a potential route to thio sugars by thermolysis or photolysis. This grant research project was completed and a final report has been received.

At Southern Illinois University, Carbondale, Illinois, under a grant, the effects of temperature and of different alcohols upon the hydrogen chloride-catalyzed alcoholysis of amylose triacetate and \underline{D} -glucose pentaacetate to form alkyl glucosides were investigated. Most of the alcoholyses produced 3:1 to 4:1 ratios of α - to β -glucosides at 30° C., 55° C., or at reflux. A synthetic route to alkyl α -glucoside was provided by the direct removal of β -glucoside with an anion-exchange resin. A homologous series of methyl terminal 4-deoxymaltooligosaccharides was prepared. A terminal 4-deoxy group did not block β -amylase action, whereas terminal 4-methoxy did. Contrary to indications in the literature, participation of a neighboring group is not advantageous in 4- \underline{O} -tosyl or 4- \underline{O} -methyl displacements.

In studies under a PL 480 grant at the Institute for Fibres and Forest Products Research, Jerusalem, Israel, the oxidation of wheat starch and waxy maize starch by bromine in acid aqueous medium was found to be a first order reaction. Pure amylose was unaffected by bromine at pH 3 and 1.7, probably because of the formation of the bromine-amylose complex.

Through extensive studies with model compounds at the Plastics Research Institute TNO, Delft, The Netherlands, a highly satisfactory method was developed for determining the position or positions occupied by the metal groups in metal starchates prepared under anhydrous conditions by previously reported procedures. The most suitable method of analysis was conversion of the metal starchates to the corresponding methyl ethers followed by hydrolysis with hydrochloric acid to glucose methyl ethers, which in turn were converted to trimethylsilyl derivatives and analyzed by gas-liquid chromatography. This work is being conducted under a PL 480 grant.

3. Starch graft copolymers. Viscosities of base-hydrolyzed starch-PAN (1:2) with an average grafted side chain (GSC) molecular weight of 780,000 (1 graft/2,400 AGU) in aqueous solution were the highest so far determined. Increasing the GSC molecular weight of PAN in starch-PAN from 286,000 to 780,000 increased the viscosity of the hydrolyzed products by 70 percent, as did increasing the grafting frequency of PAN from 4,623 to 2,435 AGU/graft when the GSC molecular weight was kept near 800,000. Starch-PAN graft copolymer (2:1) with a GSC molecular weight of 1,045,000 was prepared and hydrolyzed under a variety of conditions. Viscosities were up to 37 percent lower than those for 1:1 starch-PAN of GSC molecular weight of 800,000. Comparison of viscosities and salt tolerance of all hydrolyzed starch-PAN products /H(S-PAN) indicated that H(S-PAN) (1:1), GSC molecular weight 800,000, and H(S-PAN) (1:2), GSC molecular weight 780,000 are the most promising as water thickeners. A study showed that ceric ammonium nitrateinitiated graft polymerization of PAN onto a number of modified starches resulted in less homogeneous products.

In research completed during the year under a PL 480 grant to the Hebrew University, Jerusalem, Israel, graft copolymers of starch and polymethyl methacrylate were prepared by treating preformed methyl methacrylate

polymers with starch alkoxide dissolved in DMSO. Although considerable grafting occurred (presumably via transesterification), the reaction products were quite inhomogeneous and not judged to have much potential utility. Exploratory studies showed that the chloroformyl ester derivative of ethylene oxide prepolymers reacts with starch alkoxide to form graft copolymers. Glycidyl methacrylate was grafted to starch by anionic means to give 55-67 percent weight yields of purified graft copolymer.

4. Chemical products from starch. Direct chlorination of low D.S. cyanoethylated starch under alkaline conditions produced products whose pastes have favorable viscosity properties and high clarity for potential use as a pigment coating adhesive. Paper coated with clay containing this type of adhesive had high wax pick values.

Resorcinol-formaldehyde-treated starch as a reinforcing agent in two types of styrene-butadiene-rubber (SBR), nitrile, and natural rubber has been studied in detail. In SBR 2105 the best tensile properties were achieved with 45 phr starch and 8 percent resorcinol-formaldehyde based on starch.

Survey studies were made to determine the utility of various other formaldehyde condensation products, in addition to resorcinol-formaldehyde, for treatment of starch to obtain reinforcing agents. Highly reactive phenol bases, pyrogallol and catechol were as effective as resorcinol-formaldehyde but are expensive. Low-cost bases were not reactive enough. A wide variety of resol-type resins, particularly phenol-formaldehyde and melamine-formaldehyde, could be employed to treat starch for improved reinforcement.

Resorcinol-formaldehyde also functioned as an additive to zinc starch xanthate, giving about double the reinforcement of the latter when used alone in paper. There was no interference with the xanthate crosslinking reaction in coprecipitation. Paper treated with starch polyethylenimino thiouran was studied to determine its ion exchange (acid and base) capacity; it was found to be stable over a short range of repeated cycles. Several polyamines that are low-molecular-weight analogs of polyethylenimine have been found to be gel formers in reaction with starch xanthate.

Urethane plastics, obtained by pressure-molding mixtures of starch, castor oil, and pp'-diphenylmethane diisocyanate, showed a decrease in tensile strength after 1 week of aging in 10-percent NaOH, 3 percent sulfuric acid, or hot water (65° C.). The use of modified polyols instead of castor oil increased tensile strength and resistance to hot water, alkali, and acid. The polyols were obtained by heating castor oil with glycol glycosides, sorbitol, or ethylene glycol in the presence of a catalyst.

Contract research completed during the year at the Institute of Paper Chemistry, Appleton, Wisconsin, showed that starch xanthides added to paper pulp furnishes are adsorbed by processes involving electrostatic interactions Sorption activation energy of starch xanthide prepared from D.S. 0.08 starch xanthate was found to be nearly the same as that from D.S. 0.12 starch

xanthate. Pulp consistency had little effect upon xanthide sorption if the alum concentration was maintained between 2 X 10^{-4} and 2 X 10^{-5} M. Over this range, alum concentration influences xanthide retention and effectiveness more than does fiber surface area or the sorption temperature.

At the Ahmedabad Textile Industry's Research Association, Ahmedabad, India, the preparation of copolymers of cereal starches with polysaccharide gums such as guar, locust bean gum, and gum arabic was investigated. Mixtures of starch and these gums in several ratios were heated at various temperatures and times with different amounts of hydrochloric acid catalyst, and the products were isolated and characterized. The products obtained from starch and gum guar were evaluated at the Northern Division as deflocculant additives in papermaking. To date none of the products shows promise for this use. These studies, conducted under a PL 480 grant, were completed during the year.

In other research under a PL 480 grant to this institution, a commercial sample of hydroxyethylated corn starch of degree of substitution of 0.1 was characterized, and the distribution of hydroxyethyl groups in the anhydroglucose monomer unit of the starch was determined. The results indicated that the C-2 position of the anhydroglucose unit was etherified to the extent of 84 percent. Most of the remaining ether groups were located at the C-6 position with negligible reaction at C-3. These findings were corroborated by periodate oxidation. A new crystalline compound, anhydro mono-Q-hydroxymethyl-D-glucose, was isolated and characterized.

- 5. High-amylose (HA) starch film. Viscose solutions, prepared and blended with various proportions of alkali-dispersed HA starch, produced films with good clarity and flexibility. However, values for tensile strength, percent elongation, and MIT fold test were lower than those for cellophane. Film prepared from HA starch acetate of low D.S. showed improved flexibility when starch granules surviving acetylation were disrupted. Gelatinization of the HA starch acetate in an autoclave appears to be the most practical method among several tested for obtaining a material that will give a more flexible film at low relative humidities.
- 6. Thermal reactions of starch. In studies under a grant to the University of Pittsburgh, Pittsburgh, Pennsylvania, when cornstarch was pretreated with a 1:1 acetone-water wash to limit foaming, levoglucosan was obtained in 43 percent crude yield in a batch process run in a tubular reactor with ordinary electric heating. Two continuous reactors with screw-type conveyors were designed for vacuum operation. The 1-inch diameter reactor with a combination of electric and dielectric field heating, gave 30 percent yields but clogging was encountered. A 4-inch reactor is being constructed with flexible agitator blades to alleviate the problem.

At the University of Edinburgh, Edinburgh, Scotland, investigations on thermal degradation of starches revealed the following results for

differential thermal analysis (DTA) of starches and maltodextrins:
(1) Simple salts markedly affect the thermograms; (2) differences in thermograms of common starches are small and hence DTA cannot be used to characterize various starches; (3) the initial endotherm for amylopectin was lower than that for amylose; (4) the physical structure of the starch granule had the greatest importance in determining thermogram properties; and (5) thermograms of maltodextrins are similar to those of starch, with thermal stability increasing with chain length of oligomer. This work is being conducted under a PL 480 grant.

C. Microbiology and Fermentation

1. ARS culture collection. During 1967, 1,377 molds, 598 yeasts, and 612 bacterial cultures were distributed. Of these, 2,044 were supplied to domestic investigators and 543 were sent abroad. Twenty-five named cultures of Alternaria were acquired from the Quartermaster Depot Laboratories and 22 from the American Type Culture Collection for use in the antibiotics screening program. Several samples of tall fescue were collected from a Missouri pasture in which cattle showed signs of fescue foot toxicity. A large number (201) of isolates of fungi were placed in the temporary collection for further examination. (See also "Forage Utilization - Northern Region" RPA 407.)

Satisfactory conditions for sporulation of breeding stocks of Hansenula holstii were established. Both temperature and composition of the sporulation medium were critical. Through the use of genetically marked strains, evidence was uncovered for the presence of two independent complementary sets of spore-lethal factors in the breeding stocks. Segregation of three auxotrophic markers were followed in two independently derived diploids. In each, segregation of the nutritional markers relative to one another and to viable and nonviable spores were inconsistent from one sporulation to another. These diploids exhibited unique morphological changes during vegetative growth. Numerous stalked cells bearing a terminal bud appeared in young populations. Segregation ratios of auxotrophic markers in clones derived from the terminal bud and from the parent cell of these stalked forms differed markedly. The locus controlling agglutinating cell types of H. wingei segregated with that determining mating type in each of 600 tetrads analyzed. Various intercrosses and backcrosses have been started to establish cell lines which consistently produce either strongly or weakly agglutinating progeny. Production of an extracellular agglutination inhibitor is apparently determined by a single genetic factor.

Contract research by the American Type Culture Collection, Rockville, Maryland, revealed that the 104 strains of fungi previously reported have now survived a total of 30 months of storage at liquid nitrogen temperatures; 99 were suspended in 10 percent glycerol and 5 in 10 percent DMSO.

Initial grant research efforts at Baylor University, Houston, Texas, have been directed toward gaining an insight into organisms of the genus

<u>Hansenula</u> and toward developing techniques for detailed study of their cytology. Experiments have dealt with definition of growth of haploid and diploid strains under varying conditions, characterization of sporulation of diploid cells, and the correlated phase-contrast microscopy of both. Initial attempts to prepare cells for electron microscopy show promise.

Fungus and <u>Streptomyces</u> strains known to produce high yields of citric, fumaric, and gluconic acids; penicillin and streptomycin antibiotics; and proteolytic and amylolytic enzymes were investigated. A strain producing fat was also studied. For each microorganism, spores derived from both lyophilized and unlyophilized preparations were used to start appropriate fermentations. Statistical analysis and comparison of the results of hundreds of such fermentations indicated very definitely that lyophilization did not result in selection of lower yielding strains. Hence, lyophilization of industrially important cultures is a safe method to maintain strains without deterioration of their ability to form desirable products. This research, completed during the year, was conducted at the University of Allahabad, Allahabad, India, under a PL 480 grant.

In the past year 14 streptomycete cultures, with supporting taxonomic data for each, have been submitted by the Central Drug Research Institute, Lucknow, India, in accordance with the provisions of a PL 480 grant. All have been tested at the Northern Division for their ability to produce antibiotics. Eight of the cultures demonstrated activity against a grampositive bacterium (Bacillus subtilis); five against a grampositive bacterium (Agrobacterium tumefaciens); and seven against a mold (Mucor ramannianus). In addition, one was particularly active against yeasts. All of the cultures have been added to the ARS Culture Collection for use in future screening programs.

Sixty streptomycete cultures, with taxonomic data and electron micrographs for each, have been submitted by the National Institute of Agronomic Research, Madrid, Spain, as provided by a PL 480 grant. A total of 33 of the cultures have been tested at the Northern Division for their ability to produce antibiotics. Activity was demonstrated by 12 against a gram-positive bacterium (Bacillus subtilis); by 3 against a plant pathogenic gram-negative bacterium (Agrobacterium tumefaciens); and by 11 against a mold (Mucor ramannianus). All of the cultures have been added to the ARS Culture Collection for use in future screening programs.

2. Studies on enzymes. Enzymic reactions are being investigated to evaluate possibilities of their use to prepare chemical products for industrial applications.

A routine method was developed to obtain glucose-6-phosphate dehydrogenase by stepwise elution chromatography on hydroxyapatite. Overall recovery is 23 percent with 30-fold purification. Glucaric acid dehydratase, purified ninefold by precipitation with $(NH_4)_2SO_4$ and chromatographed on DEAE-cellulose columns, showed only 50 percent of its activity after being chromatographed. Transglucosyl amylase has been tentatively shown by electrophoresis and carbohydrate tests to be a glycoprotein. Gluco-amylase can be immobilized on DEAE-cellulose at pH 4.0 in 0.05 M sodium acetate. A dilute (1%) starch solution passed through a column with the suspended enzyme shows glucose formation equivalent to hydrolysis by the free enzyme. Apparent activity of the bound enzyme is about one-sixth that of the free enzyme.

By isolation of the corresponding peptide dimers after a series of enzymic degradations, two types of crosslinkages were demonstrated in Micrococcus lysodeikticus cell wall glycopeptide. In one, pentapeptide monomers were linked through N-(D-alanyl)-lysine. This link is hydrolyzed by Myxobacter ALI protease. Pentapeptide monomers of the second dimer were linked through the D-alanyl-L-alanine linkage, split by Streptomyces ML endopeptidase. A Streptomyces exo-N-acetyl hexosaminidase was found which acts as both an exo- and endo-β-N-acetylmuramidase.

Attention was turned to wall peptidoglycans which contain α,α' -diaminopimelic acid (DAP) and the stereochemistry of linkages involving this component. In <u>Bacillus megaterium</u> KM, the <u>meso</u> isomer of DAP predominates and the amino groups on the <u>L</u>-carbons are involved in peptide linkages to glutamic acid residues. While most of the amino groups on the <u>D</u>-carbon are free, some are substituted and probably crosslink peptide subunits since they are liberated by a new <u>Streptomyces</u> endopeptidase. Minor amounts of <u>D</u>, <u>D</u>-DAP are present. These residues have no free amino groups; endopeptidase treatment does not liberate these groups. This research is conducted under a PL 480 grant to the University of Liege, Liege, Belgium.

- 3. Fermentation with spores. Three Aspergillus species, A. wentii, A. flavus, and A. oryzae, were shown to have strong invertase activity. Preliminary studies showed spores of A. oryzae capable of metabolizing galactose to three new metabolites whose identities are under investigation. Low levels of tetracycline added to spore suspensions effectively controlled bacterial contamination without interfering with metabolic activity. A new method of column fermentation was developed in which fungal spores were either trapped in a resinous matrix or crosslinked with a synthetic polymer. Substrate flow rate was affected by the ratio of spores to suspending matrix and the percent conversion by the concentration of substrate and column height. The same column was still active after standing for 3 weeks at room temperature.
- 4. <u>Biological insecticides for Japanese beetle</u>. A lytic enzyme, contained in culture supernates of a <u>Bacillus</u> sp., P-1167, was found to lyse vegetative cells of <u>B. popilliae</u> but not affect the sporangium of spores or prespores. Availability of this lytic enzyme affords the means for purifying spore preparations produced <u>in vitro</u>. Sporulation on solid media has been

increased to about 20-30 percent and standard conditions yield about 5 X 107 spores per ml. Sporulation in liquid medium containing carbon has been increased to 1.5 X 10⁶ spores per ml. Effective sporulation in both solid and liquid media depends upon inocula prepared with minimum vegetative transfer from the spore stage. Work has been initiated to study the hemolymph lipids of healthy and diseased grubs. This is the only major component of hemolymph not as yet examined.

Contract studies at the University of Minnesota, Minneapolis, Minnesota, on the transfer of genetic determinants of sporulation have been completed. Final results indicate that four separate gene elements covering two proteolytic enzymes, an antibacterial factor, and competence are associated with the gene controlling spore formation in Bacillus subtilis. "Controller" genes also are indicated. Genetically controlled nutritional deficiencies also influence sporulation. Transformation of Bacillus cereus was not possible. Competence for uptake of extracellular DNA was not detected under any condition tested with a variety of DNA species including that from B. cereus phage. These phages were isolated and characterized. No evidence for an episomal DNA regulating sporulation was detected. Data indicated the presence of an inhibitory material in vegetative cultures of Bacillus popilliae which could be partially removed by dialysis or growth in a biphasic system. In addition, an unidentified nutrient was supplied by associated growth of a Rhodotorula yeast. Presporulation or early sporulation events occurred when B. popilliae was grown in association with Rhodotorula in a biphasic culture system.

In contract research at Baylor University, Houston, Texas, cells from hemolymph in early stages of infection were fixed immediately upon collection and examined by electron microscopy. Vegetative cells and prespores prepared in this manner are indistinguishable from vegetative cells of laboratory cultures. In confirmation of NU observations, a large proportion of vegetative cells at all stages of infection appear granular or light by phase microscopy and are largely devoid of cytoplasm by electron microscopy. Thus, most vegetative cells in the hemolymph apparently do not sporulate and die. The 20 percent maximum sporulation obtained in colonies may be about the same as that in the larvae and may be all that can be expected in an artificial culture system for producing spores. Research work under this contract has been completed.

Techniques, based on label distribution in metabolized products from ¹⁴C substrates, have been developed under a contract at Michigan State University, East Lansing, Michigan, for following the biochemical changes which occur during growth and sporulation of sporogenic and asporogenic strains of <u>Bacillus popilliae</u> on solid media. Further study of the occurrence and properties of catalase, a heme enzyme, in spores and refractile bodies was undertaken. With NRRL B-2309M, addition of whole blood prior to autoclaving caused up to 36 percent sporulation on a solid medium. This finding is contrary to results of earlier studies by NU's Fermentation Laboratory with blood and blood components. Labeled barbiturate has been shown to be incorporated into cellular RNA, but the pathway is not known.

- 5. Preparation of microbial polysaccharides. Under a grant to Cornell University, Ithaca, New York, continued studies on cells of yeasts and bacteria coated with positively charged colloidal aluminum oxide show that the cells can be readily coagulated by addition of uncoated cells. The coagulation was more efficient than that obtained with cationic polymers ordinarily used for this purpose. The presence of phosphate has been found to be of critical importance in the flocculating process. This work is part of a study to find new fermentation techniques that could lead to increased yields in production of microbial polysaccharides.
- 6. <u>Fermentation acids</u>. Research on fermentation acids is in progress under PL 480 grants at several foreign institutions.

At the University of Lodz, Iodz, Poland, a comprehensive program of selection of mutants of <u>Aspergillus terreus</u> for superior strains with which to produce itatartaric acid has been completed. Some mutant strains do, indeed, produce considerably more itatartaric acid than the parent strains, but the yield is still too low to be commercially practical even though a thorough study was undertaken to find the best medium and physical environment for acid production.

It has been found that itatartaric acid exists in aqueous solution in equilibrium with β -hydroxyparaconic acid in nearly equal molar ratios. When this fact is taken into consideration, the actual yield of the itatartaric acid complex is nearly double the apparent analytical values.

At the University of Tokyo, Tokyo, Japan, five strains of <u>Acetobacter suboxydans</u> have been isolated that produce dextrorotatory tartaric acid from either glucose or 5-ketogluconic acid. A strain of <u>Sporobolomyces</u> yeast and a <u>Brevibacterium</u> strain were also found to produce dextrorotatory acid, but only from 5-ketogluconic acid. A study of cultural conditions of two strains of <u>A. suboxydans</u> succeeded in increasing yields of tartaric acid somewhat.

The pathway to tartaric acid appears to be from glucose, through gluconic, 5-ketogluconic and glycolic acids, to tartaric acid. 2-Ketogluconic acid is a byproduct not utilizable for tartaric acid formation. A program is underway to find mutants that are unable to produce 2-ketogluconic acid and would then presumably produce greater yields of tartaric acid.

In other research at the University of Tokyo, <u>Endomycopsis fibuliger</u> IAM 4347 (now NRRL Y-7069) was selected by extensive survey for production of mevalonic acid. Maximum yield was 1,000 µg per ml. in 4-5 day aerobic fermentations at 30° C. The culture medium contained 10% glucose, 0.5% polypeptone, 0.1% yeast extract, 0.5% KH₂PO₄, 0.3% NH₄Cl, 0.05% MgSO₄·7H₂O, and 1% CaCO₃. Although not required, calcium pantothenate often stimulated mevalonate production as did KCN added on the fourth day. In addition, two crystalline antibiotics (brefeldin A and varrucarin A) inhibitory to

animal viruses were isolated from soil fungi. The antibiotics are effective against Newcastle disease virus strain Miyadera, herpes simplex strain HF, and vaccinia virus. Plague-inhibition tests using chick embryo fibroblast cell monolayers and tube culture assays were used. Although these substances are known antibiotics, their antiviral activity has not been reported before. The chemotherapeutic index of brefeldin A using HeLa cells is approximately one and that of verrucarin A is two. This research was completed during the year.

- 7. Plant antibiotics. Chromatographic studies of the new antiyeast antibiotic produced by Alternaria oleracea show it to be different from known available antibiotics. Screening of 160 additional Alternaria strains uncovered 94 more species with antimicrobial activity. One of several antibiotics produced by A. brassicicola was isolated and partially characterized. Nuclear magnetic resonance and spectral studies indicated the presence of a hydroxyl, a nitrile, and several methylene groups as well as considerable unsaturation. The elemental formula was determined as $^{\text{C25H30011N}}$ with a molecular weight of 540. It was optically active with a rotation $^{\text{C27D25}}$ + 20.1. Biological tests showed activity against some molds and bacteria but not against yeasts. It remained stable when autoclaved at 121° C. This project was discontinued during this reporting period.
- 8. Beta-carotene synthesis. Under a PL 480 grant to the Agricultural University, Poznan, Poland, the following factors that affect β-carotene production by mated cultures of Blakeslea trispora were studied:
 (a) Replacement of citrus molasses by spent mycelium; (b) use of spent mycelium as an adjunct to the medium; (c) stimulatory activity of various microbial cells. Aqueous extracts of the mycelium and the organic acid fraction of these extracts did not enhance carotene yields. Variously prepared digests of spent mycelium enhanced yields only to the same extent as untreated spent mycelium. Spent mycelia could partially replace both the cottonseed embryo meal and citrus molasses, thereby lowering the cost of the fermentation medium in the process.

D. Technology--Process and Product Development

1. Cereal xanthides and xanthates. Increasing the number of thionocarbonate groups in xanthide-treated paper failed to improve the wet strength of handsheets. A new laboratory method of incorporating xanthide was found which increased wet-tensile values for handsheets from 2,200-2,500 to 3,300-3,900 meters without increasing the amount of starch xanthide required. Aging tests on the process revealed that paper made with starch xanthide showed no significant loss in sulfur content during storage for 30 days.

The considerable quantities of data obtained in machine trials of cerealderived products on the 32-inch paper machine are being processed by computer. A pilot-plant apparatus for continuous production and application of starch xanthides has been constructed and used in evaluations of process variables. As wet-end additives, starch xanthate-polyethylenimine reaction products are equal to or better than commercial cationic starches of high quality and, in addition, provide wet strength.

2. Starch derivatives for protective coatings. In contract research at Battelle Memorial Institute, Columbus, Ohio, exposure of the exterior paint series for 18 months indicates some mildew growth, particularly on those paints containing dibutyl tin dilaurate. Some failure by grain cracking has occurred but no significant changes due to the use of starch derivatives have resulted. Interior exposures to north light over a 15-month period have deteriorated only to the point where discoloration is just barely visible to the naked eye. With potassium carboxymethyl starch as an additive, yellowing of the styrene-butadiene-latex paint appeared to be decreased. This result, if confirmed, could have industrial importance. Starch derivatives appear to soften interior paint films, a condition which could probably be corrected by including a small amount of zinc oxide in the formula.

Alcoholysis products were successfully prepared from dehydrated castor oil and safflower oil under a research contract at Archer Daniels Midland Company Alkyds satisfactory for evaluation were achieved by the reaction of safflower oil with tetrahydrophthalic anhydride but not with castor oil. Hypophosphorus acid was an excellent color retardant for both alkyd and glycoside synthesis. Drying time for alkyds, which is increased by hypophosphorus acid, has been reduced to acceptable levels by decreasing the amount of hypophosphorus acid and increasing the amount of lead napthenate drier. These modifications permitted satisfactory preparation of glycoside-based alkyds with long, medium, and short oil lengths from soybean, linseed, and safflower oils.

- 3. Protein-dialdehyde starch (DAS) glue for southern pine plywood. An exploratory mill evaluation of protein-DAS plywood glue was made under conditions more rigorous than normal owing to 39° F. ambient mill temperature at time of trial. Lower grade veneers were substituted for those scheduled because of mill problems with veneer drying. Thirty-four standard panels were prepared using typical production schedules. Quality glue bond tests were conducted by the American Plywood Association and resulted in a maximum rating of 62. A rating of 90 is required for a candidate glue subject to an approval test and an approved glue must maintain a rating of 85 in commercial production. Low moisture content in some of the veneers was indicated as responsible for erratic test results observed in this evaluation of the protein-DAS glue. Low-moisture veneers are responsible for major gluing problems encountered by the southern pine plywood industry, which currently employs phenolic resin glue.
- 4. <u>Graft copolymers</u>. Contract research on starch-acrylamide (AA), starch-acrylic (AAc), and starch acrylonitrile (AN) grafts at General Mills, Minneapolis, Minnesota, has shown all of the graft copolymers except AN to

have greater water solubility and lower gelatinization temperatures than ungrafted starch. All of the graft copolymers effectively thicken 15-percent sodium hydroxide solutions and retain good viscosity during storage for several months. In HCl only AA grafts hydrate and show potential as thickeners. None hydrate or disperse in an electrolyte such as 10 or 25 percent NaCl. Fractionation studies showed all products from AA or AN grafts to be heterogeneous. AN from pilot preparation appeared to contain a large amount of ungrafted starch as compared to that prepared in the laboratory. Upon alkaline hydrolysis, however, both formed extremely viscous solutions indicating that composition and molecular weight may vary considerably without producing gross changes in the properties of the hydrolysis product.

- 5. Starch for rubber reinforcement. Under a research contract, scientists at the University of Akron, Akron, Ohio, prepared SBR masterbatches containing up to 100 phr of starch xanthide incorporated by oxidative coupling with sodium nitrite. Starch xanthide accelerated vulcanization, gave large increases in rubber modulus, and reinforced more effectively than zinc starch xanthate. A variety of starch xanthate-rubber coprecipitation procedures have been developed for practical production of starch reinforced masterbatches.
- 6. Microbial polysaccharides. In studies on continuous fermentation, a 25-gal. single-stage fermentation for production of B-1459 was run for 13 days. Steady state conditions were maintained for 10 days, during which the daily yield was 81 percent based on glucose. The overall yield was about 75 percent. Fermentation cost of this continuous process, including further improvements in medium, is estimated at 16 cents vs. 19 cents per pound for the 48-hour batch fermentation.

Studies on kinetics of batch fermentation enabled reduction in fermentation time from 48 to 32 hours. Fixed capital investment is reduced by 20 percent and cost of product is decreased by 1.2 cents per pound.

RPA 702 - PROTECT FOOD SUPPLIES FROM HARMFUL MICROORGANISMS
AND NATURALLY OCCURRING TOXINS

C. Microbiology and Toxicology

1. Aflatoxin investigations. Numerous organisms were examined for a fungal system capable of catalyzing aflatoxin transformations. Several fungi are capable of transforming about 50-60 percent of aflatoxin B₁ into a compound having 1/20th as much toxicity, but none was found which was capable of total degradation of aflatoxin. A fluorescing compound separated by silica gel chromatography from an aflatoxin M sample could be aspertoxin. This new mycotoxin was recently reported to have been found in Aspergillus flavus. Cultures capable of degrading commarin are being further screened for mutants with the ability to degrade aflatoxin.

In contract research at the University of South Dakota, Brookings, South Dakota, <u>Aspergillus ochraceus</u> and <u>A. sulphureus</u> strains were grown on soybeans and wheat and checked for toxicity on chicks and mice. The <u>A. ochraceus</u> group has the largest number of strains to date which produce mycotoxins on wheat and soybeans. Many toxic strains were found in the <u>A. fumigatus</u> group, but deaths in toxicity tests on mice were attributed to lung abcesses, congestion, pneumonia, and enteritis rather than to liver damage. Low toxicity was found in the <u>A. niger</u> group and the <u>A. restrictus</u> group.

- 2. Reduction of viable microorganisms in corn dry-milled products. Highest microbial populations in corn dry-milled products were in the flour, degerminator fines, and feed fractions. Three methods for effective microbial control are (1) treating corn before processing by steaming for 1 minute, heating at 200° F. for 20 minutes, or treating with hot solutions of sanitizing agents; (2) heating the degerminator streams at 150° F. for 1 hour at 15-20 percent moisture before milling; or (3) treating the dry-milled product after milling at 200° F. temperature for 10 minutes.
- 3. Microbiological processes and products. The Pioneering Laboratory for Microbiological Chemistry conducts a long-term program of basic investigations on the metabolic processes of microorganisms and on the metabolites they elaborate. In studies on linkages in the cell walls of spiral and rodshaped bacteria, spiral walls were found to be the more extensively crosslinked. The regulation of lipid methylations into N-methyl, cyclopropane and methyl-branched groups is being investigated with methionine mutants of Agrobacterium tumefaciens. Zymonic acid (C₆H₆O₅), first isolated at the Northern Division in 1950, has been shown to be produced by 16 species of yeasts from 13 different genera. Eleven crystalline azo derivatives of zymonic acid have been prepared and their chemistry has been investigated. An unidentified yeast (NRRL Y-6954) has been found to excrete 3-D-hydroxypalmitic acid in copious amounts. The substance, occurring on cell walls of one mating type of a yeast, which causes sexual agglutination with the other mating type, was studied. Each active site in this substance is a protein, stabilized by one disulfide bond. Structures of no other sexually agglutinative substances are known in this much detail.

RPA 901 - ALLEVIATE SOIL, WATER AND AIR POLLUTION

B. Chemical and Physical Investigations to Improve Products

l. Biodegradable surfactants and detergent builders. A series of unsaturated di- and tricarboxylic acids were reacted with hypochlorous acid and epoxidized with alkali. These epoxy intermediates failed to react appreciably with starch in granule form. The chlorohydrin of aconitic acid readily hydrolyzes to dihydroxytricarballylic acid. This compound is a good sequestering agent for calcium and magnesium in alkaline solution and will be studied as a builder for detergents.

These studies are supplemented by a contract with Ashland Chemical Company, Minneapolis, Minnesota, for research on surfactants from polyol glycosides and by a grant to Western Michigan University, Kalamazoo, Michigan, for investigation of homogeneous catalytic oxidation of starch. Since this work has only recently been initiated by these institutions, significant results have not yet been obtained.

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RPA 406 - NEW AND IMPROVED FOOD PRODUCTS FROM FIELD CROPS

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RPA 407 - NEW AND IMPROVED FEED, TEXTILE AND INDUSTRIAL PRODUCTS FROM FIELD CROPS

Industrial Products

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 - RPA 702 PROTECT FOOD SUPPLIES FROM HARMFUL MICROORGANISMS
 AND NATURALLY OCCURRING TOXINS

Microbiology and Toxicology

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WHEAT UTILIZATION (Northern Region)

Northern Utilization Research and Development Division, ARS

USDA and Cooperative Programs

	- :	Scien	tii	st Ma	n_1	Vaars	FV	7 1068		
Location and Nature of		:Scientist Man-Years FY 1968: Research Problem Area								Total
Intramural Work	:			407					:	10001
Peoria, Illinois	:		:		:		:		:	
	:		:		:		:		:	
Food	:		:		:		:		:	
A. Chemical Composition and	:		:		:		:		:	
Physical Properties	:	8.0	:		:		:		•	8.0
B. Microbiology and Toxicology	•	0.7	•		:	6.3	:		:	7.0
C. TechnologyProcess and	:		:		:		:		:	
Product Development	:	1.5	:		:		:		:	1.5
	:		:		:		:			
	:		:		:					
Industrial Products	:		:		:		:			
A. Chemical Composition, Physi-	:				•					
cal Properties and Structure	:		:	1.1	:		:		:	1.1
B. Chemical and Physical Investi-	:		:		:		:			7.1
gations to Improve Products	•		:	13.7	:		:	1.4	:	15.1
C. Microbiology and Fermentation	:		:	6.4			:	12.4		6.4
D. Technology—Process and	•		:	O. T			:			0.7
Product Development	:		•	4.5	:		:		•	4.5
Total		10.2	;	25.7	:	6.3	:	1.4	:	43.6

Intramural program is supplemented by extramural support representing (a) 0.5 SMY's at State Agricultural Experiment Stations , and (b) 11.4 SMY's at other U.S. institutions .

- 1/ RPA 407: 0.2 (Chemical Composition, Physical Properties and Structure) RPA 702: 0.3 (Microbiology and Toxicology)
- 2/ RPA 406: 2.0 (Microbiology and Toxicology)
 RPA 407: 8.5 (Chemical Composition, Physical Properties and Structure,
 1.9; Chemical and Physical Investigations to Improve
 Products, 2.7: Microbiology and Fermentation, 1.3;
 Technology--Process and Product Development, 2.6)

RPA 702: 0.2 (Microbiology and Toxicology)

RPA 901: 0.7 (Chemical and Physical Investigations to Improve Products)

Problems and Ubjectives

The dominant factor in the wheat economy of the United States continues to be a production capacity that can outpace U.S. and foreign demand. Research on wheat seeks to solve the problems hindering the development of markets for the full productive capacity of U.S. agriculture. The Northern Division research seeks to find new industrial markets for wheat, particularly in the Pacific Northwest where corn is not grown, and to improve yield and quality of wheat flour for use in foods.

Major objectives of current research are to develop and evaluate alternate ways to:

- 1. Convert wheat flour and starch to chemical products whose use will be economically advantageous to selected segments of the paper industry.
- 2. Improve yield and quality of wheat milling fractions.

Progress - USDA and Cooperative Programs

RPA 406 - NEW AND IMPROVED FOOD PRODUCTS FROM FIELD CROPS

A. Chemical Composition and Physical Properties

- l. Microscopic and ultrastructure of wheat. Protein films, formed on water surfaces and stained with osmic acid, show small lipid bodies which range in size from 0.25 μ to 0.025 μ . The abundance of these lipid bodies suggests that they may have originated from the oil of the aleurone cells or the germ and were introduced during milling. Electron microscope observations on patterns of the spread protein have not revealed any significant differences between good and poor quality hard and soft wheats. Small particles were observed which form beaded strands when protein is fixed and stained with uranyl acetate. Their size, about 300 Å in diameter, can be related to the molecular weight of native gluten and suggests a value in the neighborhood of 5-8 million.
- 2. Characterization of wheat gluten proteins. Comparison of glutenins from different varieties of wheat showed that proteins from better quality varieties are precipitated from solution by salt more easily than proteins from poorer quality wheats in the same class. The salt precipitation technique, which permits comparison of both type and amount of protein, may be useful in determinations of gluten quality on small amounts of flour. Starch gel electrophoresis of reduced and alkylated glutenins disclosed some differences among varieties representing different classes but there are few distinct differences among good and poor quality varieties in the same class. Carbohydrate content in the glutenins ranged from 3 to 18 percent, with hard wheats generally containing less than soft wheats. The solubility of glutenin proteins was improved by reduction and

S-aminoethylation. Fractional precipitation of aminoethylglutenin produced material especially rich in glycine and tyrosine and poor in histidine, methionine, valine, and cysteine. This material, which accounts for 22 percent of glutenin has a larger molecular volume than purified gliadin. A quantity of purified gliadin sufficient for an extended study on conformation was prepared from Ponca wheat flour.

- 3. Milling characteristics. Contract studies, completed during the year at the University of Nebraska, Lincoln, Nebraska, show that milling properties and protein content of wheat can be controlled by (1) crossing soft, high-protein wheats with hard; low-protein wheats and selecting crosses having desired qualities, and (2) treatment of wheat with enzymes having specificity for pectin or protein. Starch gel electrophoresis of reduced and alkylated glutenin fractions indicated qualitative differences among gluten proteins of 10 different varieties of wheat. Differences were greatest among varieties representing different classes and least among varieties in the same class. Ten distinct differences were apparent in glutenins from good and poor qualities in the same class. This result suggests that small structural differences in glutenins may produce substantial changes in quality.
- 4. Other relevant research. Research on wheat starch is integrated with that on corn starch. Results are reported under "Corn Utilization" RPA 406, Section A.

B. Microbiology and Toxicology

- 1. Rennin-like enzyme. The rennin-like enzyme produced by R. oligosporus was found to be stable at 40°C. or below and at pH of about 6. At pH below 2 or above 7, its activity was rapidly destroyed. When R. oligosporus was grown in a milk medium, the organism produced a compound which inhibited growth of the cheese-starting culture. The inhibitor is presumably a glycoprotein and can be separated from the milk-clotting enzymes by gel filtration.
- 2. Other relevant research. Research on wheat is integrated with similar studies on corn. Results are reported under "Corn Utilization," RPA 406, Section C.

C. <u>Technology--Process and Product Development</u>

1. Milling and fractionation. Pressure treatment of hard red winter wheat grains at three levels of elevated moistures prior to standard milling increased break flour yield, decreased reduction flour yield, and lowered the amount of coarse residue left in fractionation by air classification. It increased the protein content and reduced the ash content of patent flour. Caustic peeling of hard red winter wheats resulted in reduced response to milling and air-classification procedures. Yields of the finer fractions were lower with a corresponding increase in coarse residue.

RPA 407 - NEW AND IMPROVED FEED, TEXTILE AND INDUSTRIAL PRODUCTS FROM FIELD CROPS

Industrial Products

A. Chemical Composition, Physical Properties and Structure

- 1. Chemistry of glycoprotein linkages. In grant research at Marquette University, Milwaukee, Wisconsin, novel approaches to the synthesis of glycosides of hydroxyamino acids were successful. The production of <u>DL</u> serine-<u>O</u>-xyloside in 30-percent yield from 2-nitroethanol xyloside indicates an economical route to large-scale syntheses of serine-<u>O</u>-glycosides. An <u>O</u>-xyloside of serine has been synthesized in 30-percent yield by the condensation of <u>N</u>-benzyloxycarbonylserine <u>p</u>-nitrobenzyl ester with 2,3,4,-tri-<u>O</u>-acetyl-<u>D</u>-xylosylbromide, followed by hydrogenation. The kinetics of hydrolysis of serine, threonine, and hydroxyproline-<u>O</u>-glycosides in acidic and alkaline solutions has been elaborated.
- 2. Fluidization of flour. Experiments with stirred fluidized beds, conducted at Iowa State University, Ames, Iowa, under a grant, revealed that the final percentage of expansion was greater for deeper beds. Comparisons of flours containing a fluidizing agent with starch and some other industrial powders demonstrated that flours respond moderately well to stirring in a fluidized bed. An equation has been developed for predicting diffusion rates of water in potato starch, giving an insight into its physical structure. The thermal conductivity of starch was found to be too high to be a limiting factor in most heat—exchange situations.
- 3. Other relevant research. Research on wheat starch is integrated with that on corn starch. Results are reported under "Corn Utilization," RPA 407, Section A.

B. Chemical and Physical Investigations to Improve Products

1. Chemical modification of wheat gluten. Reduction of disulfide bonds of wheat gluten and other proteins by NaH in DMSO requires at least 2 moles of NaH, and sulfinylmethyl carbanion apparently does not reduce sulfide bonds. Preliminary photochemical studies with cereal proteins indicates that radiation cleaves disulfide bonds and modifies aromatic amino acids. Labeling studies with radioactive DMSO conclusively prove that photolysis in DMSO results in incorporation of DMSO into the protein backbone. Halogenated acetic acids were found to add to alkyl vinyl ketones. This appears to be a novel organic chemistry reaction having potential synthetic utility.

Under grant research at the University of Chicago, Chicago, Illinois, a number of strained, highly reactive cyclic esters containing sulfur and phosphorus were synthesized. These include 2-hydroxy-\alpha-toluenesulfonic acid sultone, the corresponding 5-nitro sultone, 4-nitro-catechol cyclic sulfate, and catechol cyclic phosphate. Kinetic studies on the reaction of

imidazole and N-methyl-imidizole with 2-hydroxy-5-nitro- α -toluenesulfonic acid sultone in deuterated aqueous solutions indicate that the imidazole nitrogen acts as a general base in these solvent systems. In contrast, the imidazoles participate in nucleophilic displacement reactions with the sultones in nonaqueous solvents such as dimethyl sulfoxide.

- 2. Other relevant research. Research on wheat starch is integrated with that on corn starch. Results are reported under "Corn Utilization," RPA 407, Section B.
- C. Microbiology and Fermentation
- D. Technology--Process and Product Development

Research in these categories is integrated with similar investigations on corn. Results are reported under "Corn Utilization," RPA 407, Sections C and D.

RPA 702 - PROTECT FOOD SUPPLIES FROM HARMFUL MICROORGANISMS
AND NATURALLY OCCURRING TOXINS

B. Microbiology and Toxicology

- 1. Reduction of viable microorganisms in flour. Strains of actinomycetes (552) and bacteria (145) isolated from wheat flour have been lyophilized and added to earlier collections to form a basis for future classification and fermentation studies on flour-inhabiting microorganisms.
- 2. Other relevant research. Research on aflatoxins and related studies pertinent to wheat utilization are integrated with similar studies on corn. Results are reported under "Corn Utilization," RPA 702.

RPA 901 - ALLEVIATE SOIL, WATER AND AIR POLLUTION

E. Chemical and Physical Investigations to Improve Products

Research in this category on wheat is integrated with similar studies on corn. Results are reported under "Corn Utilization," RPA 901.

Publications - USDA and Cooperative Programs

RPA 406 - NEW AND IMPROVED FOOD PRODUCTS FROM FIELD CROPS

Chemical Composition and Physical Properties

Seckinger, H. L., and Wolf, M. J. 1967. Lipid distribution in the protein matrix of wheat endosperm as observed by electron microscopy. Cereal Chem. 44(6), pp. 669-674.

Microbiology and Toxicology

Hesseltine, C. W., and Wang, H. L. 1967. Traditional fermented foods. Biotechnol. Bioeng. 9(3), pp. 275-288.

Technology -- Process and Product Development

- Kent, N. L., and Evers, A. D. (Research Association of British Flour-Millers, St. Albans, England). 1968. Milling of solvent-extracted wheat, semolina and flour. I. Effect on endosperm fragmentation and protein shifting. J. Sci. Food Agr. 19(1), pp. 20-30.*
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RPA 407 - NEW AND IMPROVED FEED, TEXTILE AND INDUSTRIAL PRODUCTS FROM FIELD CROPS

Industrial Products

Chemical Composition, Physical Properties and Structure

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^{*}Research supported by PL 480 funds.

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Chemical and Physical Investigations to Improve Products

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RPA 702 - PROTECT FOOD SUPPLIES FROM HARMFUL MICROORGANISMS AND NATURALLY OCCURRING TOXINS

Microbiology and Toxicology

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WHEAT UTILIZATION (WESTERN REGION)

Western Utilization Research and Development Division, ARS

USDA and Cooperative Program

		Scie	nti	st Man-	Year	rs F.Y.	196	8
Location of Intramural Work	:_	R	ese	arch Pr	oblo	em Area	:	
	:	601	:	406	:	407	:	Tota1
	:						:	
California (Albany)	:		:		:		:	
Chemical Composition and Physical	:		:				•	
Properties	:	3.0		5.0	:	0	:	8.0
Flavor		0		4.0	:	-	:	4.0
Color, Texture and Other Quality			:		:		:	
Factors	:	0		0	:	-	:	0
Microbiology and Toxicology		0	:	3.0	:	0	:	3.0
Technology Process and Product			:		:		:	
Development	:	14.6	:	2.0	:	2.0	:	18.6
Total		17.6	:	14.0	:	2.0	:	33,6

Intramural program is supplemented by extramural support representing (a) 1.6 SMY's per year at State Agricultural Experiment Stations , (b) 2.4 SMY's per year at other U.S. institutions, and (c) P.L. 480 funds in 4 countries representing 54,625 U.S. dollars equivalent per year.

^{1/} RPA 601 - Chemical Composition and Physical Properties 1.2 SMY; Color, Texture and Other Quality Factors 0.4 SMY.

^{2/} RPA 406 - Chemical Composition and Physical Properties 0.7 SMY; Flavor 0.6 SMY; Color, Texture and Other Quality Factors 1.1 SMY.

Problems and Objectives

The dominant feature of the wheat economy in the United States is a production capacity that can outpace consumption. Research on utilization of wheat seeks to solve the problems hindering the development of markets for the full productive capacity of U.S. agriculture. The emphasis is on expanding overseas dollar markets for U.S. wheats; developing new wheat food products for long-term market development in food-short nations abroad; raising the domestic consumption of wheat foods by increased variety, quality, and convenience; finding means to upgrade wheat millfeeds to recover fractions of nutritious food quality; and developing new and improved feeds from wheat.

Major objectives of the research are to develop and evaluate alternative ways for:

- 1. Improving baking properties of bread flours by controlled maturation acceptable in European countries.
- 2. Maximizing utilization of U.S. winter wheats in blends with weaker wheats in European countries.
- 3. Developing protein-enriched export foods from wheat flours and millfeed fractions and concentrates.
- 4. Improving quality of continuous-mix baked goods.
- 5. Improving quality retention in frozen femmented bread doughs.
- 6. Developing processes to enhance the nutritive value of wheat for ruminant, poultry and swine feeds.

Progress - USDA and Cooperative Programs

RPA 601 - EXPANSION OF FOREIGN MARKETS FOR U.S. PRODUCTS

A. Chemical Composition and Physical Properties

1. Mechanisms of Wheat Flour Maturation. Extraction of lipids from flour with petroleum ether somewhat decreased the amount of hydrogen sulfide released in a subsequent mixing of dough under nitrogen. Treatment with methyl bromide also decreased the subsequent release of hydrogen sulfide. These results suggest that the protein purothionin may have a role in maturing mechanisms.

Under a research grant, the University of Wisconsin is following up observations that peroxidase activity in wheat plant tissues rises following infection with plant pathogens in an attempt to determine the biological

role of peroxidase. The polymerization of proteins caused by peroxidase is also caused by tyrosinase. The polymerization can be induced with polylysine and does not seem to involve sulfhydryl groups or disulfide bonds.

Oxidative gelation of wheat-flour pentosans involves an intermolecular cross-linking of the chelate-bridge type of ferulic acid ester groups which can form coordination complexes with adventitious calcium ions. Ligand structures were probably hydroxyquinones. Confirmatory evidence is being sought by The Swiss Federal Institute of Technology, Switzerland, supported by P.L. 480 funds. A kinetic study indicates that oxidation of caffeic acid by peroxidase in the presence of hydrogen peroxide involves the direct introduction of one or more additional hydroxyl groups into the aromatic nucleus, followed by further oxidation to hydroxyquinones.

These studies provide basic information currently needed for improving baking properties of bread flour by controlling maturation.

2. Protein Changes During Malting. Contract research on the changes that occur in wheat proteins during malting has been concluded at the University of Minnesota. Protein Efficiency Ratios (corrected to casein at 2.50) for samples of eight wheat varieties ranged from 0.85 to 1.36 (mean 1.14) in rough inverse correlation with protein content. The PER's for the same wheats malted at 12° and 15° C. were not significantly different from those of starting wheats. Supplementation with lysine at 0.1, 0.2, and 0.3% addition produced an equivalent response in malted and unmalted wheat samples. Test animals showed no preference for either malted or unmalted samples. The animal feeding studies showed that no significant increase in nutritional value of wheat occurs as a result of sprouting (malting); however, these results by no means eliminate the potential of the method as a valuable first step for simplified processing of wheats.

This study provides basic information currently needed for the development of protein enriched export foods from wheat.

- B. Flavor (no current research)
- C. Color, Texture, and Other Quality Factors
- 1. Wheat-Bran Pigments. In contract research at Oregon State University, colored substances extracted from red and white brans were found to have more similarities than differences. Benzene or ethanol extracted material containing 3.6 to 7.6% nitrogen from red and white wheat brans. Dioxane-water extracted highly colored material with 3.8 to 12.0% nitrogen and molecular weights of 20,000-50,000. The presence of arginine, aspartic acid, glutamic acid, glycine, leucine, phenylalanine, proline, and three-nine indicates a peptide or protein material. Polyphenolic substances also occur. Oxidation of the colored material produced fragments with 2,4- and 3,4-hydroxylated benzoic or cinnamic structures.

This study provides basic information currently needed for the development of protein enriched export foods from wheat.

- D. Microbiology and Toxicology (no current research)
- E. Technology--Process and Product Development
- 1. Modification of Flour and Dough Tarough Maturation. Studies on the modification of flour and dough components implicated in oxidative improvement of baking performance have been initiated to provide information currently needed to improve baking properties of bread flour by controlling maturation.
- 2. Blending Capacity of U.S. Winter Wheat. Contract research to determine and quantitate the properties of U.S. hard red winter wheat flours of value in blending with weak flours to obtain optimum baking performance has been concluded. The investigators, Kansas State University, found that when flours of 12-15% protein were blended at a ratio of 1:3 with European wheat flours, definite systematic improvements in quality characteristics were observed. The improvements, per unit of protein, were equally as good with winter as with spring wheat flours. In many cases, hard red winter flours of lower protein content (9.8% and 11.2% in the wheats, 8.6% and 10.1% in the flours) also contributed noticeably to quality improvements. Obviously, the changes could not be expected to be large when added to wheats of similar protein content or in lower proportions. However, variations in test results indicate that improved tests for predicting the behavior of wheats in blends are needed to permit proper selection of wheats for export and their combination with those wheats with which they will perform best.
- 3. Changes in Flours During Bread Making. Research has been initiated to compare changes produced in specific components of winter and spring wheat flours and doughs by chemical and mechanical treatments simulating those given flours and doughs in bread-making processes as a basis for understanding the preferred positions of spring wheats in export markets.
- 4. Milk-like Products. The WU-modified Fesca Thin Mash Process to separate protein and starch of wheat flour has been successfully developed on a pilot-plant scale. Separations with a soft wheat flour were excellent over a wide range of conditions. Conditions for acceptable separation with hard red winter wheat flour were more exacting, but attainable. Solubilization of 90% of a flour (including 85% of the protein) was achieved at pH 6 with a new enzyme, Monzyme. The enzyme-treated flour is quite bland, does not coagulate or precipitate at 95° C., and may be used for preparing milk-like or other protein-containing beverages.
- 5. Heat-moisture Effects on Wheat Constituents. A study of interrelated heat-moisture effects on constituents of wheat and wheat fractions during processing has been initiated. This information is needed for the development of protein-enriched export foods from wheat.

RPA 406 - NEW AND IMPROVED FOOD PRODUCTS FROM FIELD CROPS

A. Chemical Composition and Physical Properties

1. <u>Interaction of Dough Constituents</u>. Insoluble phospholipid-metal-protein complexes formed by addition of phosphatidyl serine and Ca(II) or Ni(II) to water-soluble flour proteins contained all the tyrosine and cysteine and most of the methionine of the proteins. Complexed proteins were more acidic than the non-complexed. Blocking of sulfhydryl groups prevented complex formation most effectively when Ni(II) was the metal ion.

Modified paper electrophoresis techniques separated two acidic peptides from pepsin digests of alpha-gliadin; one fraction contained most of the aspartic acid of alpha-gliadin. This concentration of aspartic acid residues on a small fragment of the alpha-gliadin molecule suggests that charged groups are distributed unevenly on the molecular surface.

Supported by P.L. 480 funds, the Centre National d'Assistance Technique et de Recherche Applique, Antwerp, Belgium, conducted research to determine which isopropanol-soluble proteins or other protein fractions of wheat are most closely related to variations in baking quality. Protein distribution between solvents differs for ground wheat and flour. The gliadin/albumin ratio is higher for flour than for ground wheat. Results with n-butanol demonstrate the importance of lipids. Statistical studies show highly significant coefficients of correlation between sedimentation value and the amounts of proteins extracted with 40% isopropanol and 60% isopropanol. The protein fraction extracted with 40% isopropanol contains 15 to 20 components, separable by electrophoresis on starch gel. The electropherograms vary with origin of wheats, but are not changed by extraction with n-butanol. Separation on a carboxymethyl cellulose column shows the proteins extracted by 40% isoproponal contain a fraction soluble in saline acetic medium (I) and another soluble in pH 11.85 trisodic phosphate (II). The relations between I and II vary with n-butanol treatment and wheat type.

The Flour Milling and Baking Research Association, England, supported by P.L. 480 funds, found no significant difference in the binding of lipid in fat-containing doughs mixed in the same mixer at the same rate of work input to 2 and 6 Wh/lb. respectively. The high degree of scatter involved appears at least in part to be inherent in the process. Work was initiated on the distribution of lipid between gluten, starch, and water-soluble fractions of dough made from Bison (1966) and Cappelle (1966) flours. Purothionin is a mixture of components closely related to the fast moving globulins, and the crude materials contain other proteins with the electrophoretic mobilities of albumins, thus confirming the presence of a range of "proteolipids" in flour. Fractionation of purothionin yielded products each giving a single band on starch gel electrophoresis. The fast moving globulins were fractionated in the same way. Evidence of close similarity between these globulins and purothionins was strengthened by amino acid

analyses "fingerprints" and C-terminal end group determinations. Lipopurothionin has been separated into six components. Phosphatidyl choline, amino phospholipids and two glycolipids were present in the fractions. Preliminary findings have confirmed the inhibition of yeast multiplication by purothionin, but have shown no effect of this compound on gas production in yeasted doughs.

These studies provide basic information currently needed to improve the quality of continuous-mix baked goods.

B. Flavor

1. Bread Flavor. Identity was established for one of two cracker-like aroma components detectable by gas liquid chromatography in extracts of freshly baked bread. Analyses by nuclear magnetic resonance, infrared, and mass spectrometry identified the compound as 1,4,5,6-tetrahydro-2-acetopyridine. A major reaction product of proline and dihydroxyacetone, or other polyhydric compounds, the acetopyridine is stable as the bisulfite complex or as the hydrochloride. When a dilute solution was sprayed onto the crust of a loaf of stale bread, the odor of the crust appeared to become more nearly like that of freshly baked products.

Contract research has been initiated at the Evans Research and Development Corporation, New York, to study the chemical pathways involved in flavor development and deterioration in bread crust and crumb.

These studies provide basic information currently needed to improve the quality of continuous-mix baked goods.

C. Color, Texture, and Other Quality Factors

1. Character of Doughs. In contract research at Stanford Research Institute, California, the response of dough to a small sinusoidally varying shear strain was studied with a sensitive dynamic rheometer which permits the storage and loss moduli to be determined at frequencies from 0.01 to 200 cycles per second and at peak amplitudes from about 1.5 to 200 microns. Storage modulus is proportional to the energy stored and recovered per cycle; loss modulus, to the energy dissipated.) Both moduli for dough decrease markedly with an increase in amplitude. Also, when amplitude is changed during a test, about 20 minutes are required before the moduli become constant. Entrapped air (in dough) was not responsible for the amplitude dependence, as doughs mixed in vacuum behaved in the same way. With wet gluten or a 5% gelatin gel in place of dough, no amplitude dependence was shown up to 200 microns displacement. When starch was added to the gluten, amplitude-dependence again was found. Attempts to determine a yield value, indicative of a three-dimensional continuous structure, for dough indicated that if a vield value exists it is extremely small.

Supported by P.L. 480 funds, the Bread Institute of Australia used especially designed equipment to study rheological properties of dough. Tests with

laboratory-milled flours of widely differing characteristics indicated that for a highly filled system such as dough, differences in viscoelastic behavior of the gluten may be minimized so that the behavior of a dough need not necessarily reflect these differences. The influence of starch on viscoelastic behavior of different glutens at various levels of water absorption was studied. Glutens were washed from a strong high-protein flour, a very extensible biscuit flour, a well-balanced bread flour and a flour from a Hexican dwarf wheat variety. Dynamic mechanical properties were determined for various protein-starch-water compositions to study the pseudolinear region of normal doughs. In addition non-linear behavior of these three-component systems was observed. By computer analysis it appears that parameters that are related to the physical behavior of the material and are free from artifacts arising from the geometry of the cell system may be isolated from non-linear experimental observations obtained at larger deformations.

Supported by P.L. 480 funds, Osaka Women's University, Japan, is investigating the modification of physical properties of doughs and gluten by incorporation of selected polymeric materials. Farinograms of doughs containing selected reduced proteins show an increase in initial consistency after addition of iodate, followed by a prominent weakening, in comparison with farinograms of doughs with the unreduced proteins or cysteine. Extensographs show that sulflydryl compounds decrease resistance, but the resistance is recovered by the addition of iodate. Reduced polyvinyl thioglycollate, SH-ovalbumin and SH-water soluble wheat protein produced a much more desirable mellowing effect on dough than cysteine did. This effect might be brought about by the association of SH-polymer and gluten through SH-SS exchange reaction. Reduced SH-whey did not produce the same effect.

These studies provide basic information currently needed to improve the quality of continuous-mix baked goods.

D. Microbiology and Toxicology

1. Improved Quality Retention in Frozen Fermented Bread Doughs. Improvements in stability of frozen straight doughs were achieved by (a) addition of 20 to 30 ppm of potassium bromate which serves to improve volume without severely reducing gassing power, (b) selection of commercial source of compressed yeast and (c) storage of compressed yeast at 35° F. for 5 to 7 weeks prior to use, which results in an approximately additional doubling of residual activities. Selection of source of compressed yeast combined with 7-week storage of yeast at 35° F. resulted in reduced proof time, increased residual gassing power, increased residual yeast count and increased bread volume.

Active dried yeast was not as effective as compressed yeast in frezen obraight doughs.

- E. Technology -- Process and Product Development
- 1. Effects of Starch Components on Quality of Continuous-mix Baked Goods. A 3-pound batch of flour was separated into protein and starch fractions by the WU-modified Fesca Thin Mash Process. Baking tests on flour reconstituted from the fractions showed no loss of functional properties. When the process was extended to pilot-plant scale composition and yield data indicated no significant differences due to size of batch.

RPA 407 - NEW AND IMPROVED FEED, TEXTILE AND INDUSTRIAL PRODUCTS FROM FIELD CROPS

A. Chemical Composition and Physical Properties

(no current research)

B. Microbiology and Toxicology

(no current research)

- C. Technology--Process and Product Development
- 1. <u>Ruminant Feeds</u>. <u>In vitro</u> tests to evaluate digestibility of processed grains were developed. <u>In vitro</u> digestibility of popped grain was intermediate between raw grain and pressure cooked material. Microscopic studies show that starch is almost completely gelatinized in fully popped grains. A digestibility trial using sheep showed greater improvement for popped milo than for popped barley. Good gains and feed efficiency were obtained for popped milo in other sheep feeding trials.
- 2. <u>Nonruminant Feeds</u>. Bran mash before and after steam-pelleting has markedly different metabolizable energy values, but only slightly different chemical composition. Biochemical changes such as destruction of residual enzymatic activities also occur during steam pelleting. Steam pelleting causes starch gelatinization, which improves digestibility. Four previously unknown sugars in wheat bran have been found: stachyose, neokestose, fructosyl-raffinose and a tetrafructosyl glucose pentasaccharide.

The lipid and fatty acid content of seven representative samples of U.S. wheats and their millfeeds have been determined.

Publications - USDA and Cooperative Program

RPA 601 - EXPANSION OF FOREIGN MARKETS FOR U.S. PRODUCTS

Chemical Composition and Physical Properties

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Technology -- Process and Product Development

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RPA 406 - NEW AND THPROVED FOOD PRODUCTS FROM FIELD CROPS

Chemical Composition and Physical Properties

- Bernardin, J. E. and Kasarda, D. D. 1967. Ultraviolet fluorescence and absorption changes accompanying reversible aggregation of alpha-gliadin. Federation Proc. 26:820.
- Fullington, J. G. 1967. Interaction of phospholipid-metal complexes with water-soluble wheat protein. J. ipid Research 8:609-614.
- Nimmo, C. C. and O'Sullivan, Mary T. 1967. Immunochemical comparisons of antigenic proteins of durum and hard red spring wheats. Gereal Chem. 44:584-591.
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Color, Texture, and Other Quality Factors

- Tanaka, Kenji, Furukawa, Kazuyo, and Matsumoto, Miroshi. 1967. The effect of organic and inorganic acids on the physical properties of dough.

 J. Ferment. Technol. 45:566-569. 1
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^{1/} Research supported by P.L. 480 funds.

GRAIN SORGHUM UTILIZATION

Northern Utilization Research and Development Division, ARS

USDA and Cooperative Programs

				_				(0	
	Location and Nature of	:Scientist Man Years FY 1968:							
	Intramural Work	: Research Problem Area :							: Total
	3.101 -1101 1,011	:	406	:	407:	702	•	901	
		:		:	:		:		
Peori	a, Illinois			•	*				
					:				
Food				:	:				
Α.	Chemical Composition and	:			:		:		
	Physical Properties		2.9		:		:		2.9
В.	Microbiology and Toxicology		0.2			1.3			1.5
C.	TechnologyProcess and		-			-	•		
	Product Development		5.1						5.1
	11 oddo v bevezephono) · ·						.)
				۰					
Indus	trial Products			•					
A.									
£. •	cal Properties and Structure			:	0.3				0.3
В.	Chemical and Physical Investi-			:			:		
.D •	•	•		•	2.4:		•	0.4	2.8
0	gations to Improve Products			•	1.8:		:	0.4	1.8
	Microbiology and Fermentation				1.0				1.0
D.	TechnologyProcess and								7 7
	Product Development	:		:	1.1:		:		1.1
	Total	:	8.2	:	5.6:	1.3	:	0.4	15.5

Intramural program is supplemented by extramural support representing (a) 0.2 SMY's at State Agricultural Experiment Stations , (b) 3.1 SMY's at other U. S. institutions , and (c) P.L. 480 funds in 1 country representing 36,552 U. S. dollars equivalent.

- <u>I</u>/ RPA 407: 0.1 (Chemical and Physical Investigations to Improve Products) RPA 702: 0.1 (Microbiology and Toxicology)
- 2/ RPA 406: 1.4 (Chemical Composition and Physical Properties, 0.9;
 Microbiology and Toxicology, 0.5)
 - RPA 407: 1.5 (Chemical and Physical Investigations to Improve Products, 0.6; Microbiology and Fermentation, 0.2; Technology--Process and Product Development, 0.7)
 - RPA 901: 0.2 (Chemical and Physical Investigations to Improve Products)

Problems and Objectives

The growing importance of grain sorghum as a cash crop and the increasing magnitude of production challenge technology to establish a pattern of utilization that will maintain or increase the economic value of the crop. Because the bulk of the U. S. crop is grown in a relatively limited region west of the Mississippi River, there are geographic areas where freight transportation advantages should permit increased food and industrial usage of grain sorghum without significant displacement of other cereal grains. Food usage, now taking only about 1 percent of production, could be increased by improving quality, acceptability, and suitability of grain sorghum products for food. Limited industrial outlets for grain sorghum already exist. Processing techniques specifically adapted to grain sorghum should result in improved products that can maintain present industrial outlets against competition from synthetics as well as lead to increased industrial utilization.

Major objectives of current research are to develop and evaluate alternate ways to:

- 1. Get needed data on composition and processing treatments for use in making new food products from grain sorghum.
- 2. Convert grain sorghum to new or improved industrial products via technology suited to the specific characteristics of this grain.

Progress - USDA and Cooperative Programs

RPA 406 - NEW AND IMPROVED FOOD PRODUCTS FROM FIELD CROPS

A. Chemical Composition and Physical Properties

1. Chemical and physical studies on sorghum proteins. Fifteen pounds of the sorghum hybrid RS610 was debranned and degermed with a Strong-Scott barley pearler. The grits were Buhler milled to produce 65 percent first and 35 percent short flours which were defatted with n-butanol. Of many solvents tested for extracting flour protein, 70 percent ethanol solution at 60° C. was best; it extracted 40 percent of the flour protein. After milling, three hybrids (OK612, RS626, and TE77) were extracted with 60 percent t-butanol at room temperature and with 60 percent ethanol at 60° C. Either extraction removed about 40 percent of the flour protein. The t-butanol extraction was preferred because it could be freeze-dried to yield a protein soluble in electrophoretic buffer whereas ethanol solubles required dialyzing against water to obtain a difficultly soluble precipitate.

In contract research at Kansas State University, Manhattan, Kansas, six chick feeding experiments involving amino acid supplementation were completed during this period. Results indicated that both methionine and

lysine are limiting and that the amino acids in grain sorghum are readily available. Rat-feeding studies on grain sorghum showed a direct relationship between lysine content of the diets and weight gains and protein efficiency. During the 5th and 6th weeks of the feeding trials all diets were supplemented with lysine to 100 percent of the National Research Council requirement. All diets produced equal weight gains during this period with the exception of one which was low in overall protein.

2. Other relevant research. Research on sorghum starch is integrated with that on corn starch. Results are reported under "Corn Utilization," RPA 406, Section A.

B. Microbiology and Toxicology

Research on grain sorghum is integrated with related studies on corn. Results are reported under "Corn Utilization," RPA 406, Section C.

C. Technology--Process and Product Development

1. Processing studies. In the roll cooking of grain sorghum and corn grits, corn is gelatinized to a greater degree than sorghum under the same operating conditions and is more water soluble. Corn grit products exhibit slightly higher amylograph curves than do sorghum grits. If each is processed to the same degree of gelatinization, as measured by a water absorption test, amylograph curves for corn become considerably higher. Low-fat, low-ash flour can be produced by the conventional roller milling of whole grain sorghum. Experimental milling of grain tempered to 19 percent to 19.5 percent moisture produced a 50 to 53 percent extraction of sorghum flour from the first pass. This flour contained 1 percent or less fat and less than 0.5 percent ash. By remilling the shorts, the flour level approached 70 percent extraction without exceeding these fat and ash values.

RPA 407 - NEW AND IMPROVED FEED, TEXTILE AND INDUSTRIAL PRODUCTS FROM FIELD CROPS

Industrial Products

- A. Chemical Composition, Physical Properties and Structure
- B. Chemical and Physical Investigations to Improve Products
- C. Microbiology and Toxicology
- D. Technology--Process and Product Development

Research in these categories is integrated with similar investigations on corn. Results are reported under "Corn Utilization," RPA 407.

RPA 702 - PROTECT FOOD SUPPLIES FROM HARMFUL MICROORGANISMS AND NATURALLY OCCURRING TOXINS

B. Microbiology and Toxicology

Research on aflatoxins and other microbiological and toxicological studies relevant to grain sorghum utilization are integrated with similar studies on corn. Results are reported under "Corn Utilization," RPA 702.

RPA 901 - ALLEVIATE SOIL, WATER AND AIR POLLUTION

B. Chemical and Physical Investigations to Improve Products

Research in this category on grain sorghum is integrated with related studies on corn. Results are reported under "Corn Utilization," RPA 901.

Publications - USDA and Cooperative Programs

RPA 406 - NEW AND IMPROVED FOOD PRODUCTS FROM FIELD CROPS

Chemical Composition and Physical Properties

Waggle, D. H., Deyoe, C. W., and Smith, F. W. (Kansas State University, Manhattan, Kansas). 1967. Effect of nitrogen fertilization on the amino acid composition and distribution in sorghum grain. Crop Sci. 7(4), pp. 367-368.

FORAGE UTILIZATION (NORTHERN REGION)

Northern Utilization Research and Development Division, ARS

USDA and Cooperative Programs

Location and Nature of Intramural Work	:Scientist Man-Years FY 1968 : Research Problem Area 407	m , ,
Peoria, Illinois	: : : :	
Feed A. Microbiology and Toxicology	2.4	2.4
Total	. 2,4	2.4

Problems and Objectives

Tall fescue grass is grown extensively in the Southeast, in the Intermountain States, and in the Pacific Northwest as a forage crop for cattle and other domestic animals. It has excellent agronomic characteristics, producing well on marginal land and remaining green during cool weather when other grasses are dormant. Thirty-five to fifty million acres of fescue are grown for forage use in the Southeastern part of the United States alone. Cattle grazing on pasture that is predominately tall fescue sometimes develop a disease known as "fescue foot." In severe attacks, animals become emaciated and frequently die. Elimination of this disease would prevent an estimated average economic loss of about \$5 million annually.

The major objectives of current research are to discover the cause of fescue foot disease and to develop and evaluate alternate ways for its prevention.

Progress - USDA and Cooperative Programs

RPA 407 - NEW AND IMPROVED FEED, TEXTILE AND INDUSTRIAL PRODUCTS FROM FIELD CROPS

Feed

A. Microbiology and Toxicology

1. Fescue toxicity. The α-acetamido- Δ^{CB}-butenolide is toxic to cattle, but the production of fescue foot was not demonstrated. A fraction from Fusarium sp. NRRL 3249 (previously called F. nivale) grown on hay was toxic to the rabbit as was an extract of a culture grown on rice. After countercurrent distribution fractionation of the extract from the rice culture, one or possibly two compounds showed toxicity to the mouse. No butenolide was detected in the fractions. Isovaleroxy diacetoxyscirpenol, also a suspected toxin, was also undetected. A total of 171 aqueous-ethanolic extracts of cultures of molds isolated from a toxic pasture has been screened for toxicity in the mouse. Eighteen of 49 molds belonging to the genus Fusarium exhibited toxin production. One Alternaria culture also produced a toxic extract. Epicoccum nigrum produced toxins when cultured under six different conditions. At the request of the Northern Division, the University of California examined the Fusarium sp. NRRL 3249 and classified it as an atypical strain of F. tricinctum.

Publications - USDA and Cooperative Programs

RPA 407 - NEW AND IMPROVED FEED, TEXTILE AND INDUSTRIAL PRODUCTS FROM FIELD CROPS

Feed

Microbiology and Toxicology

Yates, S. G., Tookey, H. L., Ellis, J. J., and Burkhardt, H. J. (Western Utilization Research and Development Division, Albany, California). 1968.

Mycotoxins produced by <u>Fusarium nivale</u> isolated from tall fescue (<u>Festuca arundinacea</u> Schreb.). Phytochemistry 7(1), pp. 139-146.

FORAGE UTILIZATION (WESTERN REGION)

Western Utilization Research and Development Division, ARS

USDA and Cooperative Program

	:	Scientis	st Mai	n-Years F.	Y. 1968
Location of Intramural Work	: Research Problem Ar				
	:	407	:	601	: Total
	:		:		•
California (Albany)	:		:		•
Chemical Composition and Physical	*		:		•
Properties	:	1.6	•	670	: 1.6
TechnologyProcess and Product	:		•		•
Development	:	4.1	:	2.5	: 6.6
Total	:	5.7	:	2.5	: 8.2

Intramural program is supplemented by extramural support representing (a) 0.8 SMY's per year at State Agricultural Experiment Stations, (b) 0 SMY's per year at other U.S. institutions, and (c) P.L. 480 funds in 2 countries representing 91,361 U.S. dollars equivalent per year.

^{1/} RPA 407 - Technology -- Process and Product Development 0.8 SMY.

Problems and Objectives

The demand for livestock in the United States will increase 45% by 1975. Since forage crops constitute the major feedstuff for ruminant animals, the demand for forages will increase accordingly. In addition, there is an increasing demand for processed forages in European and Asiatic markets as well as in domestic markets. Fresh forage crops are the richest natural source of many nutrients for farm animals. However, up to 30-40% of the forages are lost during haymaking and ensiling. Dehydration is now the only practical means of producing products of high nutritional value in a form usable in manufactured feeds and supplements, but use of dehydrated products is restricted because of their high-fiber and growth-inhibitor content. New and improved products are needed for foreign and domestic markets.

Major objectives of the research are to develop and evaluate alternative ways for:

- 1. Improving feeds from alfalfa and other forage crops.
- 2. Improving ruminant feeds from highly lignified fibrous agricultural materials.
- 3. Developing new low-fiber products from forage crops for export markets.

Progress - USDA and Cooperative Programs

RPA 407 - NEW AND IMPROVED FEED, TEXTILE AND INDUSTRIAL PRODUCTS FROM FIELD CROPS

A. Chemical Composition and Physical Properties

- 1. <u>Unidentified Growth Factor(s)</u>. Research on unidentified growth factors in alfalfa which stimulate the growth of guinea pigs is conducted in cooperation with the University of California, Berkeley. Fractionation studies indicate that the guinea pig unidentified growth factor(s) in alfalfa is soluble in alcohol and other polar solvents. It may be precipitated from an alcohol extract by addition of acetone. Ion exchange separation of the active acetone precipitate left most of the activity in the acid fraction. Recombination of all fractions gave a growth response no greater than that of the acid fraction alone. The growth of animals on the acid fraction was better than that of animals fed the basal diet, but not as good as that of animals fed 10% dehydrated alfalfa meal. The fractionation studies for unidentified growth factors in an aqueous extract of fresh alfalfa with a microbiological assay using <u>Bacillus subtilis</u> has been completed. Adenine, adenosine, hypoxanthine, and guanosine stimulated growth of the organism.
- B. <u>Microbiology and Toxicology</u> (no current research)

C. Technology -- Process and Product Development

1. Improved Forage Products. Research on leaf-stem separation by air classification of unground dehydrated alfalfa has progressed through the pilot plant to full plant scale studies. Pilot plant and commercial plant studies have shown that losses of xanthophylls during dehydration are inversely correlated with moisture content at the discharge end of the dehydrator.

Cooperative research with the Economic Research Service has resulted in a powerful computer technique, parametric linear programming, to determine intrinsic values of experimental products and to guide process research.

A full knowledge of valuable components is necessary to evaluate the products developed from forages. Stachydrine determinations are being made on forages and feeds other than alfalfa. The major quaternary compound in all feeds tested other than alfalfa is betaine rather than stachydrine.

Pilot-scale tests of a dry process for alfalfa leaf-stem dehydration and separation produced over 300 tons of products, which are being evaluated chemically and biologically by several university and industry laboratories. Two companies are using the process.

- 2. Alfalfa Products. In contract research at the University of Nebraska, basic data on alfalfa stem digestion have been obtained from feeding alfalfa leaf-stem mixtures to sheep. In vivo improvements of digestibility of stems treated chemically were not as large as anticipated from in vitro assays. In vitro tests showed alfalfa lignin to be more resistant to breakdown than cereal straw lignin. Treatment of stems with steam has been carried out at pressures up to 600 psi, but over 300 psi there were no increased benefits. About 700 samples have been treated and assayed by the in vitro method. Some studies have been made on stem material combining chemical treatment with high pressure-high temperature. A lamb digestion trial using high pressure cooked material showed increased carbohydrate digestibility but decreased protein digestibility.
- 3. Products from Southeastern Grasses. Contract research at the University of Georgia has been concluded. Conditions necessary for the consistent production of high-quality dehydrated Coastal Bermuda grass and pearl millet were established. Carotene and xanthophyll levels were higher than in standard alfalfa meal. Carotenoid stability during storage was similar to alfalfa and similarly enhanced by ethoxyquin. Metabolizable energy for the growing chick was 1400 Kcal/kg. Xanthophyll in the grasses was equal in biological value to that in dehydrated alfalfa meal and corn gluten meal. The proximate analyses, vitamin content, amino acid composition, and mineral content for the dehydrated grasses were determined, and would be useful in computer least cost feed determination. Processing costs for dehydrating these grasses are higher than for alfalfa, but for sale in Southeastern states, this is offset by lower transportation costs.

RPA 601 - EXPANSION OF FOREIGN MARKETS FOR U.S. PRODUCTS

A. Chemical Composition and Physical Properties

Information obtained in the following investigations provides the basis for the development of products from forage crops.

- 1. <u>Estrogen Inhibitors</u>. Supported by P.L. 480 funds, The Hebrew University, Israel, has initiated an investigation of the nature of the estrogeninhibiting factors in alfalfa and other forages.
- 2. Alfalfa Saponins. Supported by P.L. 480 funds, The Hebrew University, Israel, is also studying the saponin components of alfalfa. At least nine different saponin fractions were separated from alfalfa tops and roots. A close resemblance was found between alfalfa top saponin and root saponin prepared by the same method and compared by thin-layer chromatographic separation. Hemolytic indices showed remarkably greater activity for the root saponin than from that of the alfalfa tops. Foam forming activity also differed. Soyasapogenin E was found in alfalfa, a fact that had not been previously reported. Alfalfa top saponin was harmless to Tribolium castaneum larvae while the root saponin was highly toxic. Addition of cholesterol to the diet almost completely overcame the toxic effects of the root saponin.
- 3. Natural Alfalfa Antioxidants. Research supported by P.L. 480 funds at The Experiment Station for Practical Agriculture in Milano, Italy, has been concluded. Natural antioxidants isolated from alfalfa include alphatocopherol, solanachromene and an unidentified compound RD2. Solanachromene has a lower antioxidant activity in protecting beta-carotene in inert medium than alpha-tocopherol while compound RD2 possesses greater activity. In dried alfalfa samples incubated at 37° C. for 46 days, the alphatocopherol and solanachromene do not appear to have retained protective efficiency while weak activity was still observed for compound RD2.
- B. Microbiology and Toxicology (no current research)
- C. Technology -- Process and Product Development*
- 1. Improved Forage Feed Crops. Promising results have been obtained in preliminary small-scale tests of a separation process for fresh forage. Smooth juicing rolls removed about 45% of the weight of fresh alfalfa. The juice contained 10% solids and was rich in protein, xanthophyll, carotene, minerals and vitamins. A continuous vat process separated the protein from the juice after heat coagulation. The protein coagulum can be skimmed off from the juice, eliminating the need for expensive centrifuging equipment. The material, dried by lyophylization, contained essentially no fiber and more than twice as much protein, carotene, and xanthophyll than good quality dehydrated alfalfa.

^{*} This research also has importance for RPA 407.

Publications - USDA and Cooperative Program

RPA 407 - NEW AND IMPROVED FEED, TEXTILE AND INDUSTRIAL PRODUCTS FROM FIELD CROPS

Technology -- Process and Product Development

- Guggolz, J., Herring, V. V. and Kohler, G. O. 1967. Dehydrated alfalfa composition. Correlation of Non-nutrient components with protein content. Agr. and Food Chem. 15:1052.
- Livingston, A. L., Knowles, R. E., Nelson, J. W., and Kohler, G. O. 1968. Xanthophyll loss during pilot and industrial scale alfalfa processing. Agr. and Food Chem. 16:84.

RPA 601 - EXPANSION OF FOREIGN MARKETS FOR U.S. PRODUCTS

Chamical Composition and Physical Properties

Polesello, A. 1967. The natural antioxidants in the preservation of dehydrated alfalfa. Studi in Onore Di Ugo Pratolongo, Vol. XI della Collana della rivista Agrochimica. 1/

Technology -- Process and Product Development

Taylor, Reed D., Kohler, George O., Maddy, Kenneth H., and Enochian, Robert V. 1968. Alfalfa meal in poultry feeds--an economic evaluation using parametric linear programming. Agricultural Economic Report No. 130.

 $[\]underline{1}$ / Research supported by P.L. 480 funds.

RICE UTILIZATION (SOUTHERN REGION)

Southern Utilization Research and Development Division, ARS

(RPA 406 - NEW AND IMPROVED FOOD PRODUCTS FROM FIELD CORPS)

USDA and Cooperative Program

	:	Scientist	•
Location of Intramural Work	:	Man-Years F.Y. 196	58 :
Louisiana (New Orleans)	:		:
Chemical Composition and Physical	*		:
Properties	•	3.2	:
	Total:	3.2	•

Intramural program is supplemented by extramural support representing P. L. 480 funds in 1 country representing 7,394 U. S. dollars equivalent per year.

Problems and Objectives

In the United States, the capacity for producing rice has increased faster than domestic consumption and exports. Detailed knowledge of chemical composition and physical properties of rices is needed to guide milling, processing, and product development. New and diverse food products that are economical to manufacture, convenient to prepare, and attractive in flavor and texture must be developed to increase the consumption of rice both domestically and abroad. Research is also needed to develop new and practical procedures for preventing or sealing checks and altering plasticity of rice kernels to improve milling yields.

Current objectives of this research are:

- 1. To identify the characteristics of the untreated or treated, whole or fractionated kernel that may be used to predict potential commercial uses.
- 2. To develop new and improved rice products from the flour and residual kernels prepared by deep-milling.

Progress - USDA and Cooperative Program

A. Chemical Composition and Physical Properties

- 1. Distribution of the Proteins. Intact protein bodies were isolated from rice endosperm at neutral pH and low temperature by differential centrifugation. These protein bodies readily broke down at lower pH or higher pH and high temperature. Integrity of protein bodies was maintained best with 0.5 molar sucrose in 0.1 molar phosphate buffer at pH 7.2 and 0°C. Electron microscopic examination of these organelles showed a distinct membrane at the peripheral boundary. The lamellar structure within, which was similar to the annual rings observed in growing trees, surrounded a very dense region in the center of the protein bodies. This research is being conducted under a P. L. 480 grant to Kyoto University, Kyoto, Japan.
- 2. Composition and Properties of Rice Flours and Other Products. Research is continuing on the chemical composition of high-protein rice flours prepared from six varieties of commercially milled rice and two varieties of solvent milled rice. The Satake Rice Whitener, a commercial huller, was successfully used to produce high-protein rice flour. Interrelation-ships between the nature of the milled rice and the depth of material removed affected the characteristics of the flour. Since a high proportion of chits was produced, the equipment should be modified. Although the protein content of the flour was enhanced by air-classification, the process was not efficient. In another phase of the work, tasty rice chips that have the appearance of potato chips were prepared from white rice;

preliminary tests indicate that they will store safely for three months under air in glass containers. Storage stability tests are also being conducted on a ground, extruded brown rice product, which did not become rancid at room temperature or below for six weeks.

Publications - USDA and Cooperative Program

Chemical Composition and Physical Properties

- Mitsuda, H., Yasumoto, K., Murakami, K., Kusano, T., and Kishida, H. (College of Agriculture, Kyoto Univ., Kyoto, Japan). 1967. Protein bodies in rice endosperm. Mem. Coll. Agr., Kyoto Univ., No. 92 (Chem. Ser. No. 32), pp. 17-28.1
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Technology -- Process and Product Development

- Hogan, J. T. 1967. Processed rice products. Rice J. <u>70(11)</u>, pp. 25, 28-31.
- Hogan, J. T. 1967. Southern division rice utilization research. Rice J. 70(7), pp. 91-93.
- Mazzeno, L. W., Jr., Deobald, H. J., and Hogan, J. T. 1967. High protein rice flours. U. S. Dept. Agr. ARS 72-60, 7 pp.
- Mottern, H. H., Vix, H. L. E., and Spadaro, J. J. 1967. Popping characteristics of rice. Rice J. 70(8), pp. 9, 24-31.

General

- Anon. 1968. Rice utilization research. USDA, ARS, Sou. Util. Res. and Dev. Div., 28 pp.
- Hogan, J. T. 1967. Present products and future markets. Rice Farming & Rice Ind. News 1(9), pp. 14-15.

W Emphication resulting from research under grant of P. L. 480 funds to the Foreign institution.

RICE UTILIZATION (WESTERN REGION)

Western Utilization Research and Development Division, ARS

USDA and Cooperative Program

	:	Scientist Man-Years F.Y. 1968 Research Problem Area: 406 : Total : : : : : : : : : : : : : : : : : : :	
Location of Intramural Work	:	Research Problem Area	•
	:	406	: Total
	:		•
California (Albany)	:		•
Technology Process and Product	:		•
Development	:	2.0	: 2.0
Total	:	2.0	: 2.0

Intranural program is supplemented by extramural support representing (a) 0.7 SMY's per year at State Agricultural Experiment Stations 1, (b) 0 SMY's per year at other U.S. institutions, and (c) P.L. 480 funds in 1 country representing 12,575 U.S. dollars equivalent per year.

^{1/} RPA 406 - Technology -- Process and Product Development 0.7 SMY.

Problems and Objectives

The per capita consumption of rice in the U.S. has increased about 25% over the past ten years. Most of the increase has occurred through the use of processed products featuring convenience of use, especially quick cooking rice. Most of this product is made by processes which are inefficient due to rather large losses of solubilized solids and to kernel breakage. Final product cost is therefore higher than necessary. Also all precooked rice is made from long-grain rice, which is higher in cost than medium- or short-grain rices. The need is for new and improved food products with better nutritive quality and produced at minimum increase in cost. Such products must be easy to prepare; have good texture, flavor, and appearance; and be economical to manufacture. Also needed are drastically improved milling methods.

Major objectives of the research are to develop and evaluate alternative ways for:

1. Developing new processing methods and lower cost consumer products from short- and medium-grain rices.

Progress - USDA and Cooperative Programs

RPA 406 - NEW AND IMPROVED FOOD PRODUCTS FROM FIELD CROPS

A. Chemical Composition and Physical Properties

- 1. Amylopectins of Rice. The Nagoya University, Japan, supported by P.L. 480 funds, is conducting research to determine the biochemical and physical factors in the structure of amylopectic molecules of rice starch as they relate to processing characteristics. Two major enzymic pathways appear to operate in the breakdown of carbohydrate reserve in germinating rice seeds. One is the phosphorolytic cleavage of polysaccharide (starch) by the starch phosphorylase and the other is the hydrolytic breakdown through the amylolytic enzymes (alpha- and beta-amylases). Maltose-series oligosaccharides are hardly detectable in nature; this fact casts some doubt on the second alternative in germination processes. Analysis of phosphorylase activity was initiated and evidence of phosphorylase reaction in germinating rice seeds was found. Quantitative analysis of carbohydrates indicated existence of appreciable amounts of maltose-series oligosaccharides; this suggests the existence of amylolytic breakdown of reserve polysaccharides. Studies of the functions of alpha-glucosidase and maltase were initiated. Gibberellic acid-treated rice seeds did not show marked change in carbohydrate metabolism when compared with untreated rice seeds. Adenolate protection of rice granular starch synthetase against iodoacetate inhibition was studied. Evidence suggests that the iodoacetate binds to histidine residues residing in the enzyme active site.
- B. <u>Flavor</u> (no current research)

- C. Color, Texture and Other Quality Factors (no current research)
- D. Microbiology and Toxicology (no current research)
- E. Technology--Process and Product Development
- 1. New Rice Products. Amino acid analyses of 25 samples of rices and milling byproducts showed similar compositions for all samples of each type (rice, bran, millfeed), and appreciable differences between types, as expected. Gel filtration and electrophoresis showed that a segregated major fraction of milled rice albumins is comprised of two components with similar molecular weight. Bran albumins contain more components, each in smaller proportion. Globulins of regular and waxy rice have identical components. Information obtained in this investigation is needed for evaluating potential food uses of the various products.
- 2. High-protein Rice Products. The University of California in Berkeley is conducting contract research to evaluate the cooking and processing characteristics, utility of new products, and optimum economic balance of products from white rice overmilled to various degrees. Three flours totaling 8 to 15% of the total kernels by weight were prepared by abrasively milling three consecutive surface layers from each of six varieties of milled rice. Completed analyses on the flours and on original and residual kernels include moisture, ash, protein, fat, phytic acid, and riboflavin. Cooking evaluations were made on original kernels and those remaining after each milling. Nutritive values of flours and original and residual kernels were determined by rat-feeding tests and as Protein Efficiency Ratios.

Publications - USDA and Cooperative Program

RPA 406 - NEW AND IMPROVED FOOD PRODUCTS FROM FIELD CRCPS

Technology -- Process and Product Development

Ferrel, R. E. 1968. Coated Rice. U.S. Patent No. 3,384,493.

Houston, D. F. 1967. High-protein flour can be made from all types of milled rice. Rice J. 70:12-15.

Huxsoll, C. C. and Morgan, A. I., Jr. 1968. Dicrovaves for quick-cooking rice. Cereal Sci. Today 13:203-06.

NUTRITION AND CONSUMER-USE RESEARCH

Consumer and Food Economics Research Division, ARS
Human Nutrition Research Division, ARS

USDA and Cooperative Program

		:_	Scientist Man-Years F.Y. 1968								
		:_	Research Problem Areas :								
Location of Intramura	l Work	:	701	:	703	:	704	:	708	:	Total
		:		:		:		•			
Maryland		:		:		:		:		:	
Beltsville		:	2.7	•		:	3.0	:		:	5.7
Hyattsville		:		:	1.1	:		:	1.3	:	2.4
	Total		2.7	:	1.1	:	3.0	:	1.3	:	8.1 1/

Intramural program is supplemented by extramural support representing 1.6 SMYs at U.S. institutions other than State Agricultural Experiment Stations.

^{1/} Man-years not included for basic nutrition research related to dietary carbohydrates briefly considered in this report.

Problems and Objectives

Nationwide attention is focused on the need to improve the nutritional levels of some individuals and some population groups. Nutrition education and food distribution programs are being intensified and expanded. To facilitate these programs, research must continue to add to our understanding of the nutritive values of foods, the nutritional needs of people, and how their needs can best be met by food. More effective ways must be found to inform people about foods and nutrition and to help them improve their food habits. For families and institutions, new methods of handling and preparing the continuous array of new food products must be developed so that their nutritive and other desirable qualities are retained. A continuous flow of information gained from research on food consumption and dietary levels of the population, the food management practices of families, and the factors that affect their food choices is an essential research backstop to the development and evaluation of agricultural policies and programs in food and nutrition.

Major objectives of the research are:

- 1. To determine human requirements for nutrients and foods
- 2. To assess the nutritive values in foods and develop representative values
- To evaluate food consumption patterns, dietary levels, food habits and food management practices of families and individuals
- 4. To develop and improve the procedures used by consumers to prepare and handle food so as to preserve or enhance its qualities
- To review and interpret research findings on foods and nutrition for application to action programs and to develop guidance materials

Progress - USDA and Cooperative Program

RPA 708 - HUMAN NUTRITIONAL WELL-BEING

A. Evaluation of Grain Products

1. Wheat as a protein source for adults. Wheat provided the protein needs for 12 young men in diet studies carried out under contract at Greensboro, North Carolina.

Nitrogen balance was maintained during a 75-day period in which 35 grams of protein were supplied by wheat or wheat supplemented with pinto beans, rice, or peanuts and 12 grams by fruits and vegetables. Mean plasma cholesterol, phospholipid, and glyceride levels were slightly but not significantly lower with the wheat diets than they had been before the study started. Plasma total lipid levels fell during the first 30 days and then rose gradually to levels that were slightly lower than but not significantly different from the initial levels. Plasma total essential amino acids also fell during the first 30 days of the study. By the end of 75 days, however, they had risen again to the initial levels. This readjustment back to initial levels of blood components during the last 45 days of the 75-day study indicates the need for long-term observations with human subjects when evaluating foods or food supplements for human consumption.

- 2. Utilization of wheat nitrogen. Young rats were able to utilize the amide nitrogen of glutamine less efficiently than an equal quantity of alpha-amino nitrogen from glutamic acid when these were the sole sources of nonessential nitrogen in the diet. However, no differences in nitrogen utilization were demonstrated between two groups of rats fed different levels of amide nitrogen as wheat. It would appear, therefore, that any failure to utilize part of the amide nitrogen would not limit protein synthesis when the usual wheat-containing diet is fed. Research is underway to obtain more information on the influence of age and the possible effect of calorie intake on the utilization of amide nitrogen.
- 3. Production of antibodies. Results from research carried out under a grant with Iowa State University suggest that measurements of the number of antibody producing cells in the spleen may provide a useful index of the protein quality of the diet. Although the level of antibody in the blood responds to quality and quantity of protein in the diet it appears to be a less sensitive index than the number of antibody producing cells in the spleen. There were more antibody producing cells in the spleen of rats fed a mixture of protein (3.3 percent from rice and 1.7 percent from mung bean) than when the diet contained 5 percent protein all from rice. Level of antibody in the blood was the same for both diets. When the diet contained 5 percent protein from rice and 2.5 percent from mung beans, the number of antibody cells in the spleen increased and antibody titer in the serum also increased. Results from the supplementation of rice with an amino acid mixture similar to that supplied by mung beans were similar to those observed with the protein mixture.

B. <u>Nutritional Evaluation</u> of Dietary Carbohydrates

1. Effect of dietary carbohydrates on blood lipids in man. The kind of dietary carbohydrate had little effect on blood lipids during short-term studies with 9 young men and 10 young women. The men and women ate diets in which 85 percent of the carbohydrate (40 percent of the calories) was

supplied by cooked wheat starch or by sucrose. No significant differences in serum levels of cholesterol, phospholipids, or glycerides were noted between wheat and sugar. Long-term studies in the Department's nutrition research have shown that fat and cholesterol metabolism in rats can be altered by the kind of dietary carbohydrate. The extent and manner of change varies with the strain and age of animal. The application of the results to humans is still to be established.

2. Effect of dietary carbohydrates on enzymes involved in fat metabolism. The activities of three enzymes involved in fat metabolism--glucose-6-phosphate dehydrogenase, 6-phosphogluconate dehydrogenase, and pyruvate kinase--were lower in the livers of rats fed starch than in rats fed sucrose. The enzymes also were significantly less active when the diet was high in cholesterol (25 percent dried egg) than when the diet did not contain egg. In the presence of cholesterol, carbohydrate differences were still apparent but were small in comparison with the differences observed in the absence of cholesterol. The high levels of fat and cholesterol in the livers of rats fed the egg diet do not appear to be associated with the level of activity of these enzymes.

C. Tables of Food Composition

1. B-vitamins in foods. A recently completed survey of the literature on B-vitamins in foods showed that the absolute values reported by different authors for the pyridoxol, pyridoxal, and pyridoxamine content of a specific food often differ markedly but agree quite well on the relative proportion of the three forms. Processing and storage alter the relative proportions as well as the absolute content of these three free forms of vitamin B_6 . The survey also showed that research is needed to determine the biological activity of these three forms of the vitamin, as well as the effects of processing and storage on the proportions of them in many more plant and animal products.

D. Nutritive Value of Meals as Served

1. Type A school lunch. Twenty lunch composites obtained in the fall of 1966 from each of 300 schools located in 19 states and five geographic regions were analyzed for seven vitamins. The lunches were identical with those served to sixth graders. On an average, the lunches exceeded the nutritional goal of one-third of the 1968 Recommended Dietary Allowances set by the Food and Nutrition Board of the National Academy of Sciences for 10 to 12 year olds for vitamin A, riboflavin, niacin, vitamin B_{12} and vitamin D. The average B_6 content equaled the nutritional goal. Riboflavin, which is safeguarded by the milk requirement of the type A pattern, exceeded the goal at all schools.

Meals served in some of the schools provided substantially less of some vitamins than is desirable. Vitamin A, vitamin B_6 , thiamine and

vitamin D were the nutrients most often short in lunches which failed to meet the nutritional goals. For more than half of the schools, lunches failed to meet the goal for one or more of these four vitamins. Only a small proportion of the schools served lunches that furnished less than one-fourth the daily recommended allowance for one or more vitamins. Except for this small proportion, the schools served lunches that could be considered reasonably satisfactory in vitamin content.

E. Nutrition Guidelines for Food Programs

1. Procedures for estimating the cost of food at home for the USDA food plans were revised to reflect changes in buying patterns that occurred between the 1955 and 1965 food consumption surveys. Use of the revised procedure increased the estimated costs of the low-cost and liberal food plans by 4 percent and of the economy plan by 8 percent. It resulted in no change in the estimated cost of the moderate-cost plan.

Expenditures of about 4,000 urban households surveyed in spring 1965 were compared with costs of the USDA plans at spring 1965 prices. Roughly 13 percent of the households used food valued at less than the cost of the economy plan. Twenty-five percent used food valued at less than the cost of the low-cost plan. Forty-five percent used food valued at less than the moderate-cost plan. Sixty-five percent used food valued at less than the liberal plan.

2. Nutrition research findings continue to be studied and interpreted for application to problems in food selection and food use. Special attention is given to providing support for action programs of the Department and of other government agencies. For example, the set of commodities made available to participants under the Direct Food Distribution Program was evaluated for nutritional adequacy. Also evaluated were alternative methods for improving the nutritional adequacy of the distributed commodities. These included recombination of the commodities, fortification of the commodities and the addition of new commodities. Menus using distributed commodities and based on the economy food plan are being developed for the use of leaders who work with families.

Technical assistance was given to the School Lunch Division of C&MS in the development of (1) a breakfast menu planning guide for use with the pilot school breakfast program and (2) meal patterns including minimum quantities of foods to serve, for use in special food service programs for children. An evaluation of the Type A Lunch Pattern is now being prepared using data obtained in the study of the composition of a week's lunches in 300 schools and taking into account the recently revised NRC Recommended Dietary Allowances.

RPA 704 - HOME AND COMMERCIAL PREPARATION OF FOOD

A. Food Use

- 1. Low-income families. Recipes for a broad range of commodities, including fruits, vegetables, rolled oats, cheese, nonfat dry milk, instant mashed potatoes, dry scrambled egg mix, and canned chicken were developed for use by low-income families participating in USDA food distribution programs. A pilot survey in two low-income housing developments in Washington, D.C. showed these recipes to be useful and acceptable.
- 2. National School Lunch Program. About 170 recipes were developed or reevaluated for the School Lunch Program. The recipes covered a wide range of commodities, including canned chopped meat, sweetpotato flakes, canned grape juice, frozen french fried potatoes, peanut butter, raisins, dried eggs, rolled oats, and concentrated orange juice. In addition, yield and quality information on new forms of foods has been obtained. This information on recipes, yield and quality is essential for school lunch managers who are in charge of feeding almost 19 million children in the National School Lunch Program.

B. Nutrient Composition of Wheat and Wheat Products

A study of the changes in nutrient composition of wheat during milling and processing of wheat bread products has been largely completed. Wheat flour contained little of the tocopherol and less than 30 percent of the vitamin B_6 of the whole grain. Hard and soft wheat flours retained about 25 percent of the thiamine and riboflavin of the whole wheat grain while flour from durum wheat retained about 50 percent of the thiamine and almost all of the riboflavin. In this study the durum wheats contained more thiamine than the hard and soft wheats.

The lipids of wheat grains and their flours (2.5 to 3.2 percent) were largely linoleic acid--over 50 percent of the total fatty acid content. A small amount of lipid material in the wheat grain was not extracted by solvents or solvent systems and needed an acid hydrolysis treatment to free it for analysis. This was also true of breads, cakes, and macaronis prepared from commercially milled flours. The extractable lipid from wheat and wheat flour contained more stearic acid, less palmitate, and usually more linoleate than the "bound" lipid.

Ten consumer available wheat products including breads, cereals, and doughnuts did not vary greatly in thiamine and riboflavin content among 10 cities selected as representative of a national sampling. Only 25 percent of vitamin B_6 in the whole grain was retained in baked products. Almost 50 percent of the vitamin B_6 was present as pyridoxine. Tocopherols in baked products made with vegetable shortening came mainly from the

shortening. The fatty acids were not different among regions for comparable products.

RPA - 701 - INSURE FOOD PRODUCTS FREE FROM TOXIC RESIDUES FROM AGRICULTURAL SOURCES

A. Wheat Fumigation

The storage phase of a 3-year storage study of wheat with 8 periodic fumigations has been completed. Effects upon nutrient and breadmaking qualities were investigated. Fumigants used were methyl bromide, phosphine, ethylene dichloride and carbon tetrachloride. Overall quality of wheat deteriorated during the course of the study and was reflected in the poor eating quality of bread made from the flour. Phosphine had a particularly adverse effect on the breadmaking qualities of wheat flour. Techniques and treatments for flour production may require modification should phosphine fumigation become more widely used. There was a consistent build up of inorganic bromide residue from 8 to 99 ppm as a result of methyl bromide fumigation.

B. Plant Proteins and Insecticides

Research has been initiated under contract with Wisconsin Alumni Research Foundation to investigate the biochemical and physiological response of rats to diets containing rice with or without added lysine or lysine and threonine as the source of protein. The diets will be fed with and without the addition of malathion and DDT.

RPA 703 - FOOD CHOICES, HABITS, AND CONSUMPTION

A. Food Consumption and Dietary Levels - 1965 Nationwide Survey

1. Quality of diets. Amounts of food used in U.S. households in 1965 were sufficient, on the average, to provide diets meeting the NRC Recommended Allowances. Half of the households had diets that met the allowances for all nutrients. These diets were rated "good." The other half of the households had diets that failed to meet allowances for one or more nutrients. Calcium, vitamin A value, and ascorbic acid were the nutrients most often found to be below allowances. About one-fifth of the diets provided less than two-thirds of the allowances for one or more nutrients. These diets were rated "poor."

Little difference was found in the proportion of households with diets below the allowances for one or more nutrients in the four regions--Northeast, North Central, South, and West.

Similar proportions of urban and rural households had good diets. More rural than urban diets were below allowances for vitamin A value and

ascorbic acid. But for most other nutrients studied, more urban than rural diets were below allowances.

At each successively higher level of income, a greater percentage of households had diets that met allowances. High income of itself, however, did not insure good diets. More than one-third, 37 percent, of households with incomes of \$10,000 and over had diets that were below the allowances for one or more nutrients. Almost two-thirds, 63 percent, of the households with incomes under \$3,000 had diets that did not meet allowances for one or more nutrients. Over one-third, 36 percent of the households with incomes under \$3,000 provided less than two-thirds of the allowance for one or more nutrients and rated poor. At this income level, poor diets were most frequent among urban households in the North Central and rural households in the South.

Fewer households had good diets in 1965 than in 1955--50 percent in 1965 and 60 percent in 1955. The proportion with poor diets increased over the 10 year period from 15 percent in 1955 to 21 percent in 1965. Decreased use of milk and milk products and vegetables and fruit, the main sources of calcium, ascorbic acid, and vitamin A value, was chiefly responsible for these changes in dietary levels.

Estimates of the concentration and location of households with poor diets by State and county were derived by statistical methods from the 1965 nationwide food consumption survey data in conjunction with Census statistics on population and income. A relatively high proportion of poor diets was found in the counties of the South and North Central Regions. No county had more than 30 percent or less than 9 percent of its households with poor diets. Counties with the greatest number of households with poor diets were those with large metropolitan areas. These estimates were made in response to a request by the Committee on Agriculture, U.S. House of Representatives, in their hearings on the Stennis Bill to Provide Food and Medical Services on an Emergency Basis. The estimates were published in a report to the Committee along with an evaluation of published reports on clinical and biochemical studies of nutritional status.

2. Food use by farm and urban households. Both farm and urban households have shared in changes in food consumption in recent years but farm households have made more changes than urban in their use of all the major food groups except vegetables and fruits. As a result, farm households have become more like urban households in the foods they use. In spring 1955, farm households used 19 percent more milk than urban but only slightly more (4 percent) in spring 1965. Farm households used 7 percent less vegetables and fruits in 1955; only 3 percent less in 1965. They used 33 percent less purchased bakery products per person a week in 1955, but only 18 percent less in 1965. The largest difference still exists in the

use of flour and cereals; in 1965 farm households were still using more than twice as much as urban families. Continued greater use of fats and sugars by farm families is partly related to their greater use of flour and cereals. On the other hand, farm families continue to use considerably less than urban families of soups and other purchased mixtures. Some of the shifts in food habits of farm families have resulted from the decline in their production of food for home use.

Use of convenience foods. A larger proportion of the food dollar went for convenience foods in 1965 than a decade ago. In 1955, 27 percent of the grocery bill went for 32 types of items that were classed as convenience foods. Included were all types of canned and frozen fruits, vegetables and juices; frozen, canned and dried potatoes; ground beef, frankfurters and other lunch meat; mixtures and soups; prepared flour mixes, bakery products (including purchased bread) and breakfast cereals; instant coffee; fruit ades and punches; canned and dry milk; frozen desserts; and commercially prepared puddings, pie fillings and icings. By 1965, the part of the grocery bill that went for these same items had increased to 30 percent. The percentage increase was greater for lowthan high-income families. By 1965 low-income families were spending a slightly higher percentage of their money on these convenience foods than were the higher income groups. Farm families had upped markedly the proportion of their grocery money spent on convenience foods, in part because they were buying so much more of their food and producing less.

B. Food Acceptance and Food Habits

The amounts of foods consumed and rejected by tenth grade girls and boys at four Louisiana high schools were determined from the weights of foods served and the weights left on trays on three successive days. Boys consumed more of all foods except vegetables than girls. The highest percent of waste was noted in the vegetable group (approximately 50 percent) and the second highest in fruits (30 percent). There was little waste of milk and desserts. Acceptance of certain items differed in different schools. This may have been due to different methods of preparation.

C. Nutrition Program Service

An intensified nutrition education program was initiated following release of the findings of the study on dietary levels of U.S. households surveyed in the spring of 1965. State nutrition committee chairmen were advised of the need for this program and were urged to take part in it. Nutritionists in States not having a committee were urged to form one. Assistance was provided to seven States in developing nutrition education programs. Three workshops were developed; 3 seminars and 10 talks were given to groups involved in community nutrition committees.

Bimonthly publication was continued of Nutrition Program News, which reaches some 6,000 workers in nutrition and related fields.

Publications - USDA and Cooperative Program

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Home Preparation of Food

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III. MARKETING AND ECONOMIC RESEARCH

GRAIN - MARKET QUALITY Market Quality Research Division, ARS

USDA and Cooperative Program

	: Scientist Man-Years F.Y. 1968									
Location of Intramural Work	:	Research Problem Area:								
	:	408	:	50ì	: 702	:	Total			
Georgia (Insects)	:	0.3	:		:	:	0.3			
Maryland (Beltsville)	*	1.3	:	5.4	: 1.0	•	7.7			
Kansas	:	0.5	:	1.0	0.5	:	2.0			
Kansas (Insects)		7.0	:		•	:	7.0			
Total	:	9.1	:	6.4	1.5		17.0			

Intramural program is supplemented by extramural support representing (a) 3.3 SMYs at State Agricultural Experiment Stations $\frac{1}{2}$, (b) 5.7 SMYs at other U. S. institutions $\frac{2}{2}$, and (c) P.L. 480 funds in six countries representing 269,925 U. S. dollars equivalent.

^{1/} RPA 408 1.3; RPA 501 1.0; and RPA 702 1.0

^{2/} RPA 408 0.7; and RPA 501 5.0

Problems and Objectives

Grain and cereal products are subject to damage, contamination, or deterioration in quality while in marketing channels. The estimated storage losses for the cereal grains, exclusive of insect damage, between 1951 and 1960 amounted to 92.5 million dollars annually. of this type of loss is caused by improper storage and growth of storage microorganisms. Other losses stem from injury and breakage in handling. To facilitate the marketing of grain, the industry is constantly seeking faster, automated methods of grading. Recent emphasis on consumer protection dictates that research on mycotoxins in grain used for food and feed be continued. Naturally occurring mycotoxins have been found in all the major cereal grains except oats and rye. Losses of basic grains and processed grain products in marketing channels due to stored-grain insect damage and contamination averaged 465.5 million dollars annually for the years 1951-1960, in spite of the millions of dollars expended for preventive and control measures. The projected production for 1980 is about double that of the 1950's. If losses in 1980 were reduced by one-third, a savings of about 300 million dollars would be realized not including reduced costs for improved control methods.

Major objectives of the research are to:

- 1. Develop equipment and methods to facilitate the grading of grain by automation or semiautomation.
- 2. Reduce storage, shipping and handling losses of grains.
- 3. Reduce mycotoxin production in grains and improve methods of detection and measuring mycotoxins.
- 4. Reduce losses caused by stored-grain insects through improved insect control procedures that minimize pesticide residues.

Progress - USDA and Cooperative Program

RPA 408 - QUALITY MAINTENANCE IN MARKETING FIELD CROPS

A. Quality Evaluation and Maintenance

1. Changes in wheat due to insecticides. There was no consistent or important change in the moisture content of Kansas-grown wheat treated with four inert dusts for insect protection during 36 months of storage. Treatment with the dusts reduced the grade from No. 1 or 2 to No. 3 or lower and test weight per bushel was lowered by as much as 3.4 to 4.1 pounds. However, dust treatment and reduction in test weight had no effect on the flour-yielding capacity, or on the chemical and physical

properties of the doughs or breadbaking properties. Fat acidity increased especially during the last 6 months of storage. Ash content and diastatic activity of flour made from the insecticide-treated wheats were unchanged. Other properties not affected by the dust treatment were: Sedimentation values, protein content, rheological values and initial bread-making properties. Minor decreases in loaf volume and crumb color occurred during the 36 months of storage. (FC-23)

- 2. Quality evaluation of fumigated wheat. In a 3-year cooperative study with Human Nutrition Research Division on fumigation of wheat with phosphine, methyl bromide, ethylene dichloride plus carbon tetrachloride, the stored wheat was evaluated every 3 months for various quality factors. The results indicate that the fumigated wheat was not significantly different from the controls for any quality factor studied. The only exceptions were a decrease in test weight for the phosphine-treated wheat at the 10th, 11th and 12th test periods and minor (non-significant) decreases in sedimentation values. (HN 1-22)
- 3. Quality changes due to insect-resistant treated bags. In an interdivision cooperative study to determine quality changes associated with storage of flour and cornmeal in insect-resistant treated bags the following results were obtained:

All-purpose flour - Fat acidity values increased significantly for all stored lots of flour. Some lots increased in fat acidity more than did others; the differences are believed to be related to the type of bag and amount of treatment used. There were no significant differences in bread-baking quality associated with the type of bag or insecticide treatment.

<u>Cornmeal</u> - There was no indication of significant changes in baking qualities of either regular or degermed cornmeal produced by the insecticide treatments. The only noticeable change was the increased fat acidity values of the regular cornmeal which varied somewhat due to the type of bag and level of insecticide treatment. Fat acidity of degermed cornmeal did not increase. (SP-60)

B. Prevention of Insect Infestation

1. Biology, ecology, physiology and nutrition. Certain insect progeny develop more readily in wheat that contains dockage. In a study of factors associated with this preference, wheat dockage (headhouse dust) contained 10 times more ash than clean wheat; 5 times as much fat; and 1/3 more carotene. Dust had 8% protein compared to 13% in clean wheat. Trace mineral analysis indicated that such dockage contained more Ca, Mg, Cu, Mn, Na, Fe, Zn, Ni, and Al than did clean wheat. (SP-49)

Larvae of the Angoumois grain moth did not develop in farina-bran pellets comprised of more than 30% bran or in germ-bran pellets with more than 10% bran. Newly hatched larvae failed to enter pellets containing 20% or more bran. Shortest larval-pupal periods were for moths reared in whole wheat kernels, averaging 36.1 days. The shortest larval-pupal periods of 37.3 days were in pellets of 70% farina-30% germ. The longest larval-pupal periods, averaging 145 days, occurred in 100% farina pellets. The number of larval instars increased with length of the larval period. The extremes consisted of one larva which had 12 instars in 178 days in farina while another larva molted 4 times in 20 days in whole wheat kernels. (SP-22 (A8404))

Ten insect species have developed in bulgur at 70% R.H. and 80° F. Development tooklonger than in wheat. Bulgur equilibrates at about 1% lower moisture content than wheat at the same R.H. The larger rice weevil develops faster and produces more progeny in bulgur. (SP-25 (A9353))

Surgical techniques have been developed for installing micropolyethylene tubes in the hindgut of yellow mealworm larvae for introducing and withdrawing tritiated water from the lumen to measure in vivo the rate of absorption by the rectal tissues. Results indicate rapid movement of water through rectal tissues into the hemolymph. Starved mealworm larvae held at different humidities gain weight by water absorption at R.H. above 90% and lose weight at about the same rate at lower humidities. Analysis of mealworm lipids as substrates for metabolic water production indicate lipids make up 14 to 18% of the live weight of the larvae and more than 50% are triglycerides. Major fatty acids are palmitic, oleic, and linoleic acids. (SP-27 (A9381))

The rice weevil, Sitophilus oryzae, will pick up and transmit Salmonella montevideo on the exoskeleton after being in infected wheat for 24 hours. Results to date are inconclusive concerning the presence of Salmonella in the digestive tract of the rice weevil. Preliminary and inconclusive results indicate that the progeny of Salmonella-infected rice weevils are not as easily infected internally as those from noninfected parent stock. A 1% solution of mercuric chloride was the most effective material tested to surface sterilize rice weevil and still retain Salmonella internally. One out of 200 samples of wheat from country elevators was positive for Salmonella serotype S. meuchen. had survived in wheat under normal storage conditions for prolonged periods of time--perhaps years. Most of the molds in 6 samples of wheats and 1 sample of corn were Aspergillus flavus, A. glaucus, Neurospora, Penicillium and Mucor. Bacteria included the genera Aerobacter, Flavobacterium, Achromobacter, Bacillus, the coliform group, and other unidentified groups. Molds of the Aspergillus and Penicillium groups are normal in laboratory insects. The method for extracting aflatoxin has been revised and improved. (SP-28 (A9403))

Knowledge of the biology and ecology of the lesser mealworm is needed in a study of its transmission of acute leucosis to chickens. Mealworm pupae and adults can now be sexed by morphological features. Small squares of white paper clipped together are efficient oviposition sites. Eggs hatch only within a temperature range of 70° to 100° F, but the relative humidity can range from 7 to 95%. Preoviposition for virgin females is 9 days at 80° F. Starvation induces early pupation in lateinstar larvae. (SP-35 (A9707))

The biology of the khapra beetle from India was studied with reference to diapause at various temperatures and population densities. Diapause is not a secondarily acquired adaptation of strains from colder countries. Notable tolerance to environmental stresses was noted. Proteinaceous globules in the fat body described as albuminoid granular or protein granules were numerous. Their chemical nature and function in insects is not understood, but complex cytochemical reactions and possible similarity in chemical composition to yolk spheres of insect eggs were The globules are destroyed during pupation. Quantitative studies on fat, glycogen, and protein in normal and diapause larvae showed an enormous increase of these metabolites during diapause. Fat and glycogen were the chief sources of energy during diapause. With food available, body weight, fat, and glycogen content were constant during 3 months of diapause, indicating replenishment of the metabolites by intermittent feeding. Protein levels could not be established at beginning of diapause, but the significant decrease after 9 months without food during diapause showed considerable depletion of fat and glycogen. Cytological studies of the fat bodies of starved larvae corroborated this. Other studies indicated the use of fat and glycogen as chief sources of energy during larval-adult transformation. Adults from larvae induced with food to pupate after 2 months of diapause, laid more eggs than did nondiapausing individuals. (SP-39 (A7-MQ-3(a))

Almond moths, corn sap beetles, and maize weevils were the 3 most prevalent insects found in representative samples of shelled corn stored for 8 months in a tobacco warehouse. (SP-52)

2. <u>Biological and physical control</u>. Of several biopreparations of the disease organism <u>Bacillus thuringiensis</u> tested against the Mediterranean flour moth, the "Anduze" strain from France was most effective. (SP-45 (E30-MQ-1(k))

Histological study of female khapra beetle adults revealed that groups of ectodermal cells under the last abdominal sclerites near the ovipositor closely resemble glands that excrete attractive substances in Lepidoptera. Extracts of pheromones from virgin males and females of 3 ages were evaluated for attraction. If attraction is proportional to concentrations of pheromone, excretion of pheromone appears to increase with age of adults. In studies of cross-species attraction, Trogoderma granarium pheromone repelled Tenebrio molitor, Tribolium

castaneum, Dermestes maculatus, and Oryzaephilus surinamensis, but attracted Callosobruchus maculatus. Pheromone from T. molitor attracted T. granarium. (SP-41 (A10-MQ-11(k))

About 700 varieties of wheat from the World Collection were screened in India for resistance to rice weevils. Fewer than 5 weevils per test sample emerged from 2.6% of the varieties. Of 745 varieties tested against lesser grain borers, 4.8% showed resistance to attack. Of varieties screened against both species, 6 were resistant to both. (SP-38 (A7-MQ-1(a))

The oviposition rate of <u>Sitophilus</u> <u>zeamais</u> was significantly affected by differences in varieties of sorghum grown by natives of South Africa. (SP-20 (G9177))

Laboratory experiments showed that all ages of almond moth eggs were equally susceptible to parasitism by Trichogramma evanescens. (SP-52)

3. Improved insecticidal control. Gardona, 5% malathion in a granular carbon formulation, and a low-volume malathion emulsion treatment protected wheat in small bins from insect attack for 12 months. (SP-49)

Packaged food products in contact for 7 days with inside surfaces of railway freight cars treated with a 2½% solution of dichlorvos and malathion contained dichlorvos residues of less than 0.1 p.p.m. Fumigation of empty boxcars with 165 pellets of aluminum phosphide per 1,000 ft. 3 killed 100% of test insects. Applications of malathion to newly harvested wheat by farmers using standard techniques showed wide discrepancies in amounts of insecticide actually applied—in some instances 1/50th the amount required for satisfactory insect control. Modification of application techniques will be studied. (SP-49)

Field tests on stacks of sacked delinted cotton indicated that 180 tablets of aluminum phosphide per 1,000-cubic-feet killed 100% of pink bollworm larvae exposed for 120 hours. Preliminary data from later small-scale tests indicate this dosage might be reduced considerably. (SP-50)

Amounts of malathion lethal to insects were deposited throughout corn masses in 14- and 26-foot columns of grain using thermal aerosols and forced air flow. Reversing air flow and introducing aerosols at top and bottom gave effective insect control at reduced dosage levels. Gas chromatography analysis in field-scale corn fumigations indicate that both carbon tetrachloride and ethylene dichloride applied by gravity rapidly penetrate warm corn masses. Ethylene dichloride penetrated more readily in warm corn than in either wheat or grain sorghum. (SP-50)

Two pints of 57% malathion emulsion applied to 1,000 bushels of grain sorghum as an undiluted ultra-low volume spray and as a high volume water-diluted spray gave excellent protection against insect damage. Malathion and diatomaceous earth mixed together protected the grain from insects better than either material applied separately at equivalent dosage rates. In recent field tests with $80:20~(\text{CCl}_4:\text{CS}_2)$ and $75:25~(\text{CCl}_4:\text{EDC})$, gas chromatography analysis indicated than the components separate and present a fire hazard. These two fumigants should not be recommended for use on grain sorghum regardless of temperature, unless they are recirculated. (SP-51)

Dosage rates of 2 pounds of hydrogen cyanide, 2 pounds methyl bromide, and 165 aluminum phosphide pellets per 1,000 cubic feet gave complete kills of mature and immature stages of insects in bulgur. One fumigation with HCN resulted in a residue 294 p.p.m. (allowable tolerance 90 p.p.m.). Three fumigations with methyl bromide resulted in a 56 p.p.m. residue (allowable tolerance 125 p.p.m.). Three fumigations with PH₃ resulted in a .005 p.p.m. residue (allowable tolerance 0.1 p.p.m.). (SP-25 (A9353))

Data collected from samples of shelled corn stored in tobacco warehouses showed that malathion residues decreased at a faster rate in corn stored at shallow depths than in corn stored in upright bins. Malathion residues after 8 months averaged only 3.7 p.p.m. in corn 4 to 5 feet deep, and 1.7 p.p.m. on the surface. (SP-52)

Preliminary evaluations showed that relative humidity influenced the wide range of efficacy of several inert dusts against the granary weevil. (SP-45 (E30-MQ-1(k))

4. Fate and effect of residues. It is assumed that malathion decomposition in wheat is based on enzymatic breakdown. Total lipids were determined on varieties sensitive and nonsensitive to sprouting. Total lipids decreased in both varieties until ripening and further during sprouting. This line of investigation is promising, as differences in lipid metabolism between the 2 varieties have been shown. (SP-43 (E8-AMS-6(a))

RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

1. <u>Kernel Hardness</u>. Research in Holland showed that the effect of moisture content and kernel size on the physical and biochemical properties of wheat do not parallel the effect of these factors on kernel hardness indices. At high moisture levels, the four indices of kernel hardness (resistance to cutting, resistance to compressing, Smetar value, and particle size index) indicated a decrease in kernel hardness, whereas pearling index and the Durograph Ae-value (grinding energy)

indicated the opposite. High moisture wheat gave a lower flour yield, finer granulation, lower ash content, lower protein content and lower maltose value than low moisture wheat. Kernel-size test results varied with test methods. (FC-21 (E19-MQ-1(a))

- 2. Determining Proportions of Durum and Common Wheat in Semolina Products. Research conducted in Spain showed that biochemical differences exist in Triticum durum and T. aestivum products. Generally T. aestivum contains a higher quantity of sitosteryl palmitate, digalactosyl glyceride, and/or lipoprotein. Assays based on these compounds may permit differentiation between T. durum and T. aestivum products. Also, higher quantities of a water soluble protein component were found in T. aestivum than in T. durum. (FC-22 (E25-AMS-7(a))
- 3. Quality Evaluation of All-Purpose Flour. Tolerances for the alphaamylase (falling number) content of all-purpose flour are now included in the USDA Agricultural Stabilization and Conservation Service specifications. This action was based on the work described in Marketing Research Report No. 804 entitled "Estimation and control of experimental error in the falling number test for wheat and flour." No changes in the factors studied other than falling number were found to be necessary for upgrading the quality of all-purpose flour. (FC-24)
- 4. Relation of Lipids to Grain Soundness and Quality. No significant differences were noted in the thin-layer chromatographic patterns of corn with high to normal moisture contents even though the fat acidity values decreased as the corn dried. Lipids extracted from germ-damaged wheat showed differences in the fatty acid fractions as compared to the lipids extracted from sound wheat. (FC-28)
- 5. Automatic Test Weight Device. Under contract research, an automatic test-weight device was developed which utilizes a commercial weightsensing mechanism and indicator. The sample is placed in a hopper and flows by gravity into a test kettle mounted on a scale. The automatic grain leveling, or strike-off, operation duplicates the motions of the operator's hands in manipulating the stroker for the manual grain weighing operation. This action is accomplished by a motor driven mechanism. (FC-31(C))
- 6. <u>Sound Grain Determination</u>. X-ray stereomicroradiographs of grains gave good resolution. A system for automatic scanning of the microradiographs to record damaged kernels was developed and tested. An apparatus based on vapor equilibrium for the determination of moisture in grains was developed and tested. A prototype unit

consisting of two radiation detectors and a helium neon gas laser for evaluating the optical qualities of sound and damaged grains was developed. At 80% and 90% relative humidity and 70° F, corn and wheat showed different adsorption rates for sound grain and samples with varying amounts of damaged grain. (FC-32(C))

- 7. Optical Characteristics of Grains. Spectral reflectance curves of twelve different grains were made from 230 to 2450 millimicrons and mean reflectances, variances, standard deviations, and coefficients of variation were determined. All grains gave similar reflectance curves in the ultraviolet and infrared regions. In the visible region of the spectrum, the curves for oats and barley were similar to the curves for hard red spring and hard red winter wheat. Statistical analysis of the data has not been made. Determination of the profile characteristics of the same samples of the twelve different grains was started. (FC-33(C))
- 8. <u>Isolating Germ of Grains</u>. A modified wheat slicer cut dry wheat satisfactorily but did not show promise for concentrating the germ. By use of corrugated and smooth rolls, a moderately successful method was developed to concentrate most of the germ from wheat into a small fraction. A promising short simple bleaching and drying procedure was developed. (FC-34(C))
- 9. Automatic Moisture Meter. Kernel-to-kernel moisture variations within supposedly homogeneous samples did not exhibit normal distribution characteristics and indicated a large spread in moisture content. The results of differential thermal analysis and thermal gravimetric analysis indicate that, except for soybeans, accelerated moisture removal appears feasible. Effluent analysis of conventional and microwave heated grain showed that the amount of effluents, other than water, are so small that they would not interfere in moisture determinations. (FC-35(C))
- 10. Mechanical and Automated Sampling of Grain. High speed photographs of diverter-type samplers indicate that under certain conditions of grain flow the diverters will not accept all the grain presented to their entrances and the sample obtained may not be representative. A diverter-transporter has been constructed and will be used to study how diverter design influences performance.

The performance of three gravity-type secondary samplers (Boerner, Cargo and 4-way) and three powered units (Denver, Gamet and Gamet $7\frac{1}{2}$) were studied but the data has not been analyzed statistically. In a study of barley samples drawn from 12 boxcars by mechanical samplers and a compartmented probe sampler, the samples drawn by the probe sampler were significantly higher in test weight than samples drawn by mechanical samplers. (FC-36)

- 11. Variation in Yeast and its Effect on the Experimental Baking Test. Exploratory tests indicate that variation might exist in commercial yeast purchased in the open market which will significantly affect baking tests. Experiments are being designed to determine the extent of this variation and whether we should culture our own yeast. (FC-116)
- 12. Grain Breakage Test. A Dillion Universal testing machine was found to be impractical for rapid testing. Cargill and Stein corn breakage testers were evaluated and the Stein unit was selected for further testing, using samples of corn, grain sorghum and soybeans. The results justify further use of the Stein corn breakage tester in developing a standard test. (FC-113)

RPA 702 - PROTECT FOOD SUPPLIES FROM HARMFUL MICROORGANISMS AND NATURALLY OCCURRING TOXINS

- 1. Fungal Contamination and Metabolites in Grain. A rapid technique for screening corn samples for aflatoxin was developed. It is based on the assumption that if aflatoxin is present in a sample, it will normally be found in the mechanically separated cracked corn and foreign material portion. Several samples of corn known to contain aflatoxin had about 20 times more aflatoxin in the dockage portion (30 g.) than in the nondockage portion (8 lbs.). The effect of particle size of cornmeal on the concentrations of aflatoxins was studied. Results by the visual method showed that the coarse cornmeal (retained on U. S. screen No. 14) had three to four times the concentration of aflatoxin B_1 and G_1 as did the fine meal (retained on U. S. screen No. 80). A general decrease in aflatoxin concentration is apparent as particle size decreases. (FC-18)
- 2. Aflatoxin Production in Corn. When stored, nonsterile corn was incubated at temperatures of 55° to 105°F and at moisture contents of 14.5 and 19.5%, three groups of fungi predominated. Aspergillus glaucus competed best, growing in all 10 temperature-moisture combinations. Penicillium spp. preferred the higher moisture and competed best at temperatures of 55° and 65°F but one or more species grew substantially at 94° and 105°F. A. flavus grew little or none at 14.5% moisture. It grew at 65°F at 19.5% moisture but was favored by increasing temperatures.

Relatively high levels of aflatoxin were produced at the higher moisture and temperatures with maximum production at $94^{\circ}F$. Relative amounts of aflatoxins B and G were influenced by temperature, with more B_1 and less G_1 produced with increasing temperature. Although incubation at $80^{\circ}F$ gave twice the amount of aflatoxin as incubation at $105^{\circ}F$, equal amounts of B_1 were produced and it was produced 2 weeks earlier at the higher temperature. Aflatoxin production declined after 12 weeks at 80° and $94^{\circ}F$ and after 6 weeks at $105^{\circ}F$. (FC-19(A))

Mucor, Fusarium and Penicillium grew in high moisture corn stored in the presence of low oxygen and high carbon dioxide concentrations at 15.5° and 23°C. Fusarium moniliforme, although able to grow in corn at 0% oxygen, 60% carbon dioxide and 23°C, was unable to grow at 15.5°C.

<u>Penicillium</u> can grow at about 0° C, but cannot grow at oxygen levels of 0.5% or less and it is sensitive to high carbon dioxide levels.

Fusarium moniliforme and Penicillium grew better at 23% moisture than at 28%, probably because there was less competition with Mucor and aerobic bacteria at the lower moisture content. These findings account for failures of sealed storage of 20-24% moisture corn. (FC-82(A))

- 3. Control of Microorganisms in Stored Grain. An enzyme preparation consisting of amylase and protease is being tested for its ability to preserve high-moisture grain sorghum. Laboratory storage tests using this enzyme to date have failed to produce the controlled heating and fermentation that occurs in treated farm-stored grain. At moisture contents of about 25%, treated grain has remained high In palatability and nutritive value, with an anaerobic condition existing in the grain mass. (FC-124)
- 4. Refrigerated High Moisture Corn. Shelled corn with 26-27% moisture content was placed in three insulated bins in which the initial temperatures of 70°F were reduced at the rate of approximately 5°, 17° and 30°F per day, respectively, to a holding temperature of about 30°F. Rhizopus grew abundantly at temperatures of 50° to 60°F but failed to grow in the bottom of the fast-cooled bins. Mucor was more tolerant of low temperatures and grew in nearly all of the grain. Penicillium made moderate growth in the upper (warmer) layers and became quite abundant in the middle of the fast-cooled bin after about 2 months. Fat acidity values did not increase greatly except in the upper part of the slow-cooled bins. Germination decreased most in the upper portion of the slow-cooled bins. (TF2-057B)

Publications - USDA and Cooperative Program

RPA 408 - QUALITY MAINTENANCE IN MARKETING FIELD CROPS

Quality Evaluation and Maintenance

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- Yinon, U. and Shulov, A. 1967. New findings concerning pheromones produced by <u>Trogoderma granarium</u> (Everts), (Coleoptera: Dermestidae) J. Stored Prod. Res. 3(3):251-254. (A10-AMS-11(k))
- Henderson, Lyman S. 1967. Preventing insect infestations. Proceedings Grain and Cereal Prod. Sanitation Conf.

RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

- Greenaway, W. T. and Neustadt, M. H. 1967. Estimation and control of experimental error in the falling number test for wheat and flour. USDA, MRR 804. (FC-24)
- Hart, J. R. 1967. A method for detecting mixtures of artificially dried corn with high-moisture corn. Cereal Chemistry 44(6):601-606. (FC-25)
- Johnson, R. M. 1968. Modified flour disk reflectance method as a measure of bread quality. Cereal Science Today 13(3):142-145. (FC-26)

RPA 702 - PROTECT FOOD SUPPLIES FROM HARMFUL MICROORGANISMS AND NATURALLY OCCURRING TOXINS

Tuite, J. F., Haugh, C. C., Issacs, C. W. and Huxsoll, C. C. 1967. Growth and effects of molds in the storage of high moisture corn. Journal Paper No. 3033 of the Purdue Agricultural Experiment Station. (FC-19(A))

RICE - MARKET QUALITY Market Quality Research Division, ARS

USDA and Cooperative Program

	:	: Scientist Man-Years F.Y. 1968								
Location of Intramural Work	:	: Research Problem Area :								
	:	408 :	501	:	702	: Tota	a1			
Texas	•	1.01/:	1.0	:	0.5	2.5	5			
Total	•	1.0	1.0	•	0.5	: 2.5	5			
	:	:		:						

Intramural program is supplemented by extramural support representing (a) 0.6 SMYs at State Agricultural Experiment Stations and (b) P.L. 480 funds in four countries representing 66,879 U.S. dollars equivalent.

^{1/} Insect program; Beaumont 2/ RPA 408 0.1; RPA 501 0.5

Problems and Objectives

The farm value of rice produced in the United States in 1966 was \$405,369,000. To maintain the quality of rice, precise information is needed on metabolic changes, disease organisms and insect infestation. To insure the consumer a uniform and quality product, new and improved procedures for measuring quality in the inspection, grading and quality control operations must be developed. Losses of rice caused by stored-rice insects and the cost of their control for 1951-1960 were estimated to be about \$6.45 million annually. Based on a 40% increase in production of rice by 1980, an annual savings of \$2.75 million could be realized through improved control methods which would reduce losses by 33% and the cost of control by 25%.

Major objectives of the research are to develop methods and/or equipment for:

- 1. Reducing losses of rice during storage, transportation and milling through deterioration, disease and insect infestation.
- 2. Evaluating the quality of rough and milled rice rapidly and objectively.
- 3. Detecting and reducing mycotoxins in rice in market channels.
- 4. Detecting and reducing insect infestations and pesticide residues, and lowering the cost of preventing infestation.

Progress - USDA and Cooperative Program

RPA 408 - OUALITY MAINTENANCE IN MARKETING FIELD CROPS

1. Storage Changes in Milled Rice. Additional evidence confirms that the chemical composition of the outer layer of the kernel of milled rice greatly affects its cooking behavior. Cysteine disulfhydrase activity in milled rice appears to be concentrated in the outer layer of the kernel. As storage time was increased, rice became darker, developed off-odors, had reduced tendency of cooked kernels to stick together and developed changes which are both desirable and undesirable. The nitrogen, sulfhydryl, and disulfide indices show trends associated with storage conditions and indicate significant changes in the protein material of the outer layer of the kernel. The population of microorganisms tends to decrease with time in storage. However, the proportion of Xanthomonas spp. and Aspergillus spp. increases with storage time.

(FC-73(E25-AMS-9(a))

2. Biological and Physical Control of Insects. The biological significance of the constituents of rice that attract insects is under investigation. A component of polished rice attractive to the rice weevil appears to be an acidic substance of carboxylic nature that is soluble in water. Attempts to isolate the substance have not succeeded. (SP-42 (All-MQ-3))

Following elimination of the program at Fresno, California, and before the present laboratory at Beaumont, Texas, was established just prior to this reporting year, the research on the prevention and control of insects in rice was at a complete standstill. Time has been devoted to establishing a laboratory site, erecting and equipping a building, establishing adequate supplies of insect cultures, arranging for warehouse space, and making a survey to determine exactly what and where the problems are. Sufficient progress has been made to start a new research program with this year's crop. (SP-48)

RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

- 1. Rice Sizing. Under a three-year contract, research was initiated to develop an automatic device that will separate brown and milled rice of the long, medium, and short grain varieties into whole, large broken, medium broken, and small broken kernels. No results are available. (FC-123(C))
- 2. Degree of Milling and Color of Rice. As a result of tests with the Rice Ratiospect, the Grain Division, Consumer and Marketing Service, purchased two additional instruments for critical evaluation. Preliminary results show that: (1) The instrument can be calibrated to give identical readings within an allowable tolerance; (2) compensation for error is necessary to obtain accurate measurements of degree of milling of rice containing contaminants such as heat-damaged, red rice, or chalky rice; and (3) the instrument indicates an apparent rapid deterioration (not yet defined) in rice after removal from cold storage that is not detectable by subjective measurements. (FC-91)
- 3. Hidden-Insect Detection in Grain. Techniques were developed for non-destructive detection of internal (hidden) insect infestation in grain with X-ray radiographs. A combination of proper exposure levels, film type, resulted in radiographs with sufficient detail that all stages of insect development from egg to mature larvae could be observed by microscopic examination of the film. Auxiliary chemical treatment of the infested grain has enhanced the X-ray density difference between infested and non-infested kernels but some false reactions continue to be a problem. Some progress has been made in developing a vacuum pick-up wheel for handling individual kernels of grain in an automatic X-ray scanning device. (FC-129)

RPA 702 - PROTECT FOOD SUPPLIES FROM HARMFUL MICROORGANISMS AND NATURALLY OCCURRING TOXINS

- 1. Microbiological, Chemical and Physical Deterioration of Rough Rice. The ability of strains of the Aspergillus flavus group to compete with the common storage fungi, A. chevalieri and A. candidus, was studied under storage environments conducive to deterioration. A. flavus strains infected the rice most rapidly but were gradually succeeded by the other fungi. The competitive rank of the fungi depended in part upon the ratio of spore populations placed on the rice. Strains of A. flavus produced a wide range of pigments. Their capacity to produce pigments varied among strains within a species. Variability in pigment production is a probable cause of irregularity in the development of kernel discoloration in stored rice under conditions leading to microbiological deterioration. (FC-75)
- 2. Mycotoxins in Rice. In studies carried out in Japan the chick embryo test for toxicity was tested with six highly toxic fungi (Penicillium islandicum, P. expansum, P. utrinum, Aspergillus flavus, A. ochraceus and A. clavatus). Five compounds of known mycotoxin composition were also tested. The test was positive only with aflatoxin and culture broth in which strains of A. flavus were grown. Four hundred and sixteen strains of fungi were isolated from rice and cultured broths were tested for toxicity by oral administration to mice. None of the broths were toxic. Patulin was the toxic principle produced by P. expansum and A. clavatus. The latter also produced another toxic compound presumed to be dihydropatulin. (FC-74 (All-MQ-2))

Ethylene oxide and methyl bromide controlled internal infestations of fungi when applied at a rate of 32 mg/l. However, ethylene oxide reduced seed viability to zero at application rates as low as 8 mg/l. The internal flora of rice collected and studied in India consisted primarily of <u>Aspergillus sydowi</u>, <u>A. flavus</u> and <u>A. niger</u>. Examination of grain samples from 195 storage structures in Madras, Pondicherry, Kerala and Mahaiashtra States of India showed the following results: (1) Samples from structures of wood, paddy straw, and bamboo mat revealed little evidence of fungal deterioration; (2) fungal growth was observed in samples from brick and mortar structures. (FC-98 (A7-MQ-12))

In a pilot-scale study of the development of mycotoxins in stored undried rice in the United States aflatoxin developed in a highly irregular pattern when the rice was not aerated. Concentration of the contamination was usually low but mold counts were high and characterized by a preponderance of species other than \underline{A} . \underline{flavus} strains. Competition by fungi that do not produce aflatoxin greatly reduces toxin production and accumulation. (FC-78)

Publications - USDA and Cooperative Program

RPA 408 - QUALITY MAINTENANCE IN MARKETING FIELD CRCPS

- Barber, S., Benedito de Barber, C., y Tortosa, E. 1967. Almacenamiento de arroz elaborado. III. Cambios en la composicion de los lipidos segun la localizacion en el grano. Agroquimica y Tecnologia de Alimentos 7:235-243. (FC-73 (E25-AMS-9(a))
- Barber, S., Benedito de Barber, C., Guardiola, J. L. y Alberola, J. 1967. Composicion quimica del arroz. IV. Distribucion de los azucares en el grano elaborado. Agroquimica y Tecnologia de Alimentos 7:346-353. (FC-73 (E25-AMS-9(a))

RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

- Stermer, R.A. 1968. An instrument for objective measurement of degree of milling and color of milled rice. Cereal Chemistry 45(4):358-364. (FC-91)
- Stermer, R. A. 1968. Environmental conditions and stress cracks in milled rice. Cereal Chemistry 45(4):365-373. (FC-91)
- Schroeder, H. W. and Stermer, R. A. 1967. Market quality research of rice in Texas, 1966-67. Rice Jour. 70(7):99 and 104. (FC-75 and FC-91)

RPA 702 - PROTECT FOOD SUPPLIES FROM HARMFUL MICROORGANISMS AND NATURALLY OCCURRING TOXINS

- Calderwood, D. L. and Schroeder, H. W. 1968. Aflatoxin development and grade of undried rough rice following prolonged storage in aerated bins. USDA, ARS 52-26. (FC-75)
- Kurata, H., Udagawa, S., Ichinoe, M., Kawasaki, Y., Tekada, M., Tazawa, M., Korzumi, A., and Tanahe, H. 1968. Studies on the population of toxigenic fungi in food stuffs. III. Mycoflora of milled rice harvested in 1965. J. Food Hygienic Soc. Japan 9:23-28. (FC-74 (All-MQ-2))
- Kurata, H., Tanahe, H., Kanota, K., Udagawa, S. and Ichinoe, M. 1968. Studies on the population of toxigenic fungi in foodstuffs. IV. Aflatoxin-producing fungi isolated from foodstuffs in Japan.

 J. Food Hygienic Soc. Japan 9:29-34. (FC-74 (A11-MQ-2))
- Schroeder, H. W. 1967. Milling quality of Belle Patna rice in experimental storage: A study of the effects of field fungi on subsequent invasion by storage fungi. J. Stored Prod. Res. 3:29-33. (FC-75)

FECD AND SEED - MARKET QUALITY Market Quality Research Division, ARS

USDA and Cooperative Program

	: Scientist Man-Years F.Y. 1968					
Location of Intramural Work	: Research Problem Area :					
	: 408 : 501 : Total					
	: : :					
Maryland (Beltsville)	: 1.7 : 4.6 : 6.3					
Texas	: 0.0 : 0.4 : 0.4					
Total	: 1.7 : 5.0 : 6.7					

Intramural program is supplemented by extramural support representing (a) 1.3 SMYs at State Agricultural Experiment Stations $\frac{1}{4}$, and (b) P.L. 480 funds in four countries representing 56,000 U. S. dollars equivalent.

^{1/} RPA 408 0.3; and RPA 501 1.0

Problems and Objectives

Production, processing and marketing of seeds constitutes a billion dollar industry in the United States. Expanding technology and agricultural production demands the testing of more samples and methods that will predict with greater accuracy the planting value of each seed lot. Present testing procedures are time-consuming and results frequently are not closely related to planting value of seed stocks. To consistently meet the needs of consumers, seed surpluses must be carried over from years of high production to years of low production. Seed stocks of 33 forage legumes and grasses held by dealers on June 30, 1966 amounted to 171.5 million pounds. It is known that some seed lots have a longer storage life than others but there is no method of predicting the storage life of a lot of seed. Seedborne diseases account for crop losses in the millions of dollars, yet methods for evaluating seed lots for diseases or disease organisms are impractical.

Major objectives of the research are to:

- 1. Develop improved techniques and equipment for measuring the germination, viability and vigor of commercial seed lots.
- 2. Develop improved procedures and equipment for measuring the purity and "sanitary condition" of commercial seed lots.
- 3. Develop methods and equipment for predicting the storage life of seeds.
- 4. Investigate the basic physiology and biochemistry of seeds as related to methodology and storability.
- 5. Develop methods of predicting the protein content of grains.

Progress - USDA and Cooperative Program

RPA 408 - QUALITY MAINTENANCE IN MARKETING FIELD CROPS

1. Maintaining the Viability of Seeds. Multiwall bags containing aluminum layers were impervious to atmospheric moisture. Paper bags offered little resistance to the passage of moisture. Studies on hardseededness with 24 cultivars of bean lead to recommendations that: (1) Plant breeders seek to eliminate varieties tending toward hardseededness and (2) seed distributors store varieties tending towards hardseededness under conditions which will maintain a moisture content lower than 10%. (FC-14 (E10-MQ-1(a))

2. Changes Associated with Seed Deterioration. Amylase activity of germinating barley was less from seeds with a low storage potential than from seeds with a higher storage potential and amylase decreased as the seed deteriorated.

The salt tolerance of the storage fungi, <u>Aspergillus flavus</u>, <u>A. niger</u>, <u>A. parasiticus</u>, <u>A. ochraceus</u> and <u>A. terreus</u>, was compared to that of <u>A. amstelodami</u>, a known osmophilic species. The results showed that facultative ability to live on a high salt medium exists for species of <u>Aspergillus</u> not usually considered to be osmophilic. (FC-7 and FC-136 pending)

3. Predicting the Storability of Seeds. Germination percentages of 16 seed kinds were determined at intervals up to 12 months of storage under four different conditions: 7°C-45\% R.H.; 20°C-75\% R.H.; 30°C-75\% R.H.; and open warehouse. These studies showed that the accelerated aging test was an effective means of predicting storage potential and provided useful data on the longevity of seeds under several different storage conditions. (FC-9(A))

RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

A. Germination Vigor and Dormancy

- 1. Environmental factors affecting germination. Extensive data were collected on the germination response of sorghum seed to a two-way, temperature gradient to determine efficient experimental designs and develop a method of statistically analyzing the data. Additional experiments are being conducted to resolve final problems in statistical analyses. A preliminary experiment involving Indian ricegrass seeds, indicated that temperature is probably not the principle factor controlling the laboratory germination of these seeds. (FC-5 in part)
- 2. Biochemical basis of seed germination. Studies with protein inhibitors indicated that two of the enzymes responsible for starch hydrolysis in pea seeds, alpha- and beta-amylase, are synthesized $\frac{de\ novo}{de\ novo}$ and that a third enzyme, the R-enzyme, results from the activation of a preformed enzyme. Furthermore, a particulate fraction isolated from dry seeds yielded R-enzyme activity when subjected to protolytic hydrolysis. (FC-12 (A10-MQ-6(k))
- 3. <u>Development of seed germinator</u>. An automatic syphon spray humidifier was designed, constructed and tested to replace a heater type condensate evaporator. The performance of this humidifier was

found to be more satisfactory than the condensate evaporator. A complete set of plans, specifications and performance characteristics for an alternating temperature, thermistor controlled seed germinator were prepared and published as U. S. Department of Agriculture, ARS 51-17. (FC-92 in part)

- 4. Development of a method for measuring seed vigor. Respiratory measurements during the first two hours distinguished between vigorous and non-vigorous lots of corn and barley. The uptake of C¹⁴-labelled precursors into proteins and other organic constituents of germinating seeds was positively correlated with vigor in corn, barley, and wheat. Respiration and isotope incorporation data agreed in: (1) Indicating a rapid activation of both degradative and synthetic enzyme systems in seeds during the initial hours of germination, and (2) that measurements of activities based on these systems can provide valuable insights for seed quality evaluation. Respiratory measurements also detected tomato and pepper seeds pre-treated for rapid germination. A 2-vectorial analysis of seed vigor based on seedling growth over a range of environmental conditions was developed. (FC-2)
- 5. Environmental control of seed dormancy. Light exerts two effects on the germination of bluegrass seeds. Several brief, daily exposures promote germination, but when these exposures exceed one hour they are inhibitory. Greatest inhibition occurs in the far red, particularly in the 720-730 nm region. However, red light (shorter wavelengths) is inhibitory when applied continuously. A constant temperature of 20°C is best for showing the promotive and inhibitory light effects. These results are important for applied seed testing. (FC-5 in part)
- 6. Physiological maturity and dormancy of grass seeds. Seeds of Kentucky bluegrass harvested in the ripe stage were more dormant than seeds harvested in the early or late dough stage. In contrast, seeds of orchard grass and timothy harvested in the early dough stage were most dormant. Seeds of the 3 grasses showed benefit in preventing or reducing dormancy of drying at all stages of maturity. Drying decreased hard seeds of Trifolium repens and Vicia sativa harvested in the early stage of maturity and increased hard seeds of T. repens harvested ripe. Use of potassium nitrate or prechilling the seeds in the germination medium enhanced germination of timothy seeds. Prechilling increased germination of T. repens but had only a slight effect on the germination of V. sativa seeds after storage at low temperatures. (FC-15 (E10-MQ-3(a))

7. Germination inhibitors in barley seeds. Three phenolic aldehydes previously isolated from barley were determined quantitatively using gas-liquid chromatography, paper chromatography, and UV techniques. In all cases, wild species (relatively more dormant) had higher aldehyde contents than cultivated species. When the aldehydes were applied to the same kind of seed from which they were extracted, they inhibited germination in some cases but not in other cases. More results are necessary to adequately establish the role of aldehydes as inhibitors of germination in barley. (FC-11 (A10-MQ-1(a))

B. Methods and Equipment for Purity Determination

- 1. Method for determining purity of chaffy grass seeds. In searching for suitable solvents for flotation method for purity analyses two mixtures were found which caused only negligible damage to the germination of dalliagrass seed. The two mixtures are (1) hexane-carbon tetrachloride and (2) 2, 3, dimethyl butane-carbon tetrachloride. Both mixtures can be adjusted to the proper specific gravity for flotation purity analyses of many kinds of grass seed. Limited research with an air-comparison pycnometer showed it to be useful in determining density of the mixtures. (FC-92 in part)
- 2. <u>Invisible marking of seeds for identification</u>. Organic acid-base indicators and fluorescent substances were used as invisible markers. Marking materials are visualized when treated seeds are subjected to proper pH changes or to ultraviolet light. Markers were applied dry because previous research indicated that wet applications had adverse effects on seed germination. The markers did not adversely effect the function of fungicide applied to seed. When markers and seed kind were properly matched, no detrimental effects on seed appearance, viability, or storability were observed. A number of materials are of practical use, including bivalent and trivalent iron of which the anion is a weak acid and to which an antioxidant is added. Also, some fluorescent powders were found useful. (FC-13 (A10-MQ-5(a))
- 3. X-rays and radiograms as aids in analyzing seeds. Further standardization of the X-ray photographic technique was accomplished. The following classification system was established for testing seeds for anatomical soundness by the X-ray method: Healthy (anatomically sound), mechanically damaged, empty, less developed (immature or under-developed), and insect infested. Impregnating seeds with barium chloride sometimes improves the contrast in X-ray radiograph. X-ray radiography is already becoming an established procedure for evaluating seeds in some seed laboratories and plant quarantine centers. Also, anatomical soundness is a fairly good indicator of viability in fresh seeds. Barium chloride impregnation is associated with seed viability. (FC-10 (A7-MQ-2(a))

- 4. Development of seed purity testing equipment. A new semiautomatic sample divider with built-in sample weighing device was constructed and tested for accuracy and precision. A new seed blower was constructed and placed in semiproduction line hook-up with the semiautomatic sampler and the vibrator-separator, previously developed. An exhaust hood to protect the analyst from fine dusts and fumes was completed. (FC-8(C))
- 5. Methods of determining crop varieties by seed characteristics. Soybean varieties differ in their ability to produce viny seedlings when grown under suboptimal conditions in growth chambers. The viny characteristic is modified to some extent by differences in controlled environment. The source of the protein variant, found in soybean seed by electrophoretic analysis was traced to an introduction from Manchuria called 'Mandarin.' Efforts to develop a practical test for distinguishing between annual and perennial ryegrasses based on electrophoretic patterns were not successful. The variation in protein content of individual seeds is so great that negative readings do not necessarily indicate the complete absence of the subject protein. (FC-3)

C. Seed Pathology

1. Methods of detecting and identifying seedborne pathogenic fungi. Results of studies to measure the reflectance of visible light from the conidia of eighteen fungi showed the feasibility of characterizing the spore color of Aspergillus species. Satisfactory methods for the extraction and electrophoretic separation of water soluble mycelial proteins from seedborne Aspergillus species were developed. Electrophoretic analysis of A. flavus and A. parasiticus showed the two species to have 14 protein bands in common; an indication that they may be closely related. Isolates of different species of the genus Helminthosporium were obtained and identified for use in the development of selective culture media. Certain carbon sources may exert selective effects on the linear growth of some species of Helminthosporium and Fusarium on agar. (FC-4)

The perspex freezing method was modified with the elimination of blotters. Seeds are placed in depressions formed in the perspex plates, along with a small amount of water to which Terramycin is added. Seeds are allowed to imbibe at 20°C for 24 hours and are then killed by overnight exposure to -20°C. Subsequent incubation is done at 20°C, usually under near-ultraviolet light. (FC-16 (E19-AMS-11(a))

D. Protein of Feed Grains

1. Method for predicting protein content of grain sorghums and other feed grains. A rapid colorimetric method was developed for predicting protein content in finely ground grain sorghum by substituting cupric carbonate for copper sulphate in the biuret reaction. The correlation of results between the Kjeldahl procedure and optical density readings for 77 samples of grain sorghum was r = 0.95. (FC-26)

Publications - USDA and Cooperative Program

RPA 408 - QUALITY MAINTENANCE IN MARKETING FIELD CROPS

Maintaining the Viability of Seeds

- Lowig, E. 1968. Einfluss von Samenlagerung und -verpackung auf die Keimfahigkeit und den Zerfall des Keimlings. Samenfachhandel und Pflanzenzucht. (FC-14 (E10-MQ-1(a))
- Lowig, E. 1967. Beobachtungen uber eine Form von Hartschaligkeit an Samen der Stangenbohne. Samenfachhandel und Pflanzenzucht Nr. 5. (FC-14 (E10-MQ-1(a))
- Lowig, E. 1967. Die unterschiedliche Aufnahme von Wasser durch einzelne Bestandteile des Samenkornes. Samenfachhandel und Pflanzenzucht Nr. 4. (FC-14 (E10-MQ-1(a))
- Lowig, E. 1967. Eignungstest für Samen-Lagerbehalter. Samenfachhandel und Pflanzenzucht Nr. 1. (FC-14 (E10-MQ-1(a))
- Lowig, E. 1967. Uber den Austausch von Wasserdampf zwischen eingebeuteltem Kressesaatgut und der Lagerluft. Samenfachhandel und Pflanzenzucht Nr. 3. (FC-14 (E10-MQ-1(a))
- Lowig, E. 1967. Uber den gewichtsverandernden Einflul der Warmsiegelung bei Kunststoff- und Folienbeuteln. Samenfachhandel und Pflanzenzucht Nr. 5. (FC-14 (E10-MQ-1(a))
- Marcus, A., Luginbill, B., and Feeley, J. 1968. Polysome formation with tobacco mosaic virus RNA. Proceedings National Academy of Sciences 59(4):1243-1250. (MQ 3-32)

RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

Environmental Factors Affecting Germination

Toole, V. K. and Borthwick, H. A. 1968. The photoreaction controlling seed germination in <u>Eragrostis curvula</u>. Plant and Cell Physiol. 9:125-136. (FC-5)

Biochemical Basis of Seed Germination

Woodstock, L. W., Reiss, B., and Combs, M. F. 1967. Inhibition of respiration and seedling growth by chilling treatments in <u>Cacao</u> theobroma. Plant and Cell Physiol. 8:339-342. (FC-12 (AlO-MQ-6(k))

Development of Seed Germinator

Stermer, R. A. 1968. An alternating cycle seed germinator with thermistor-controlled temperature. USDA, ARS 51-17. (FC-92)

Development of a Method for Measuring Seed Vigor

- Woodstock, L. W. and Combs, M. F. 1967. Application of the respiration test for seed vigor to pea. Proc. Assoc. Offic. Seed Anal. 57. (FC-2)
- Woodstock, L. W. and Grabe, D. F. 1967. Relationships between seed respiration during imbibition and subsequent seedling growth in Zea mays L. Plant Physiol. 42:1071-1076. (FC-2)

Physiological Maturity and Dormancy of Grass Seeds

Eifrig, H. 1967. Beobachtungen uber den Eingluss Verschiedener Lagerungstemperaturen auf den wassergehalt und die keimfahigkeit von gramineespelzfruchten unterschiedlicher reife. Saatgut-Wirtschaft Fachzeitschrift fur Samen und Saaten. Nr. 9:294-297. (FC-15 (E10-MQ-3(a))

Eifrig, H. 1967. Der einfluss von Reifestadium und Lagerungstemperatur auf die Keimfahigkeit von grassamereien. Samenfachhandel und Pflanzenzucht. Nr. 15:32. (FC-15 (E10-MQ-3(a))

Development of Seed Purity Testing Equipment

Hardin, E. E., Copeland, L. O., and Knudson, L. A. 1965. A comparison of the relative effectiveness of the boerner divider and several techniques of using the gamet precision divider. Proc. Assoc. Offic. Seed Anal. 55:140-147. (FC-8(C))

Methods of Determining Crop Varieties by Seed Characteristics

Larsen, A. L. and Caldwell, B. E. 1968. Inheritance of certain proteins in soybean seed. Crop Sci. 8. (FC-3)

Methods for Detecting and Identifying Seedborne Pathogenic Fungi

Limonard, T. 1968. Verbesserung der fliesspapiermethode fur die gesundheitsprufung von samen. Umschau in wissenschaft und Technik. 4:122-124. (FC-16 (E19-AMS-11(a))

- De Tempe, J. and Limonard, T. 1967. Some aspects of <u>Fusarium</u> infection of wheat seeds. Tech. Ber. Nr. 17:35-42. (FC-16 (E19-AMS-11(a))
- Kulik, M. M. and Combs, M. F. 1967. Carbon and nitrogen nutrition of some seedborne fungi in vitro. Proc. Int. Seed Test. Assoc. 32:591-602. (FC-4)
- Kulik, M. M. and Justice, O. L. 1967. Some influences of storage fungi, temperature and relative humidity on the germinability of grass seeds. J. Stored Prod. Res. 3:335-343. (FC-4)
- Limonard, T. 1967. Bacterial antagonism in seed health tests. Neth. J. Pl. Path. 73:1-14. (FC-16 (E19-AMS-11(a))
- Limonard, T. 1967. Some remarks on the preparation of agar tests in seed health investigation. Proc. Int. Seed Test. Assoc. 32:603-614. (FC-16 (E19-AMS-11(a))

Method for Predicting Protein Content of Grain Sorghums and Other Feed Grains

Johnson, R. M. 1968. Modified flour disk reflectance method as a measure of breadmaking quality. Cereal Chem. Today 13(3):142-145. (FC-26)

MARKETING FACILITIES, EQUIPMENT AND METHODS

Transportation and Facilities Research Division, ARS

(RPA 504 - PHYSICAL AND ECONOMIC EFFICIENCY IN MARKETING FIELD CROPS)

USDA and Cooperative Program

Location of Intramural Work	•	Commodity	:Sci	entist Man-Years F.Y. 1968
Indiana Kansas Kansas Maryland (Hyattsville) Texas		Corn Wheat Grain Sorghum Grain Rice		2.1 1.0 0.2 0.8 2.1
		Total	:	6.2

Intramural program is supplemented by extramural support representing 5.1 SMY's at State Agricultural Experiment Stations and other U.S. institutions.

Problems and Objectives

Improved varieties of many field crops with their increased yields, along with progressive cultural and harvesting practices, require improved techniques and equipment for economically handling, conditioning and storing these crops in marketing channels. Grain now must be handled and conditioned in large quantities in a short period of time yet its quality must be maintained and labor and other costs minimized. Although product deterioration in storage has been reduced in recent years due to increased acceptance of aeration, shrinkage from loss of moisture is still a factor as well as loss from handling. Physical damage to the product must be reduced to maintain a high standard of quality. Because certain field crops are susceptible to contamination by toxin-producing molds, conditioning techniques and equipment must provide mold inhibiting environments.

Major objectives of this research are to:

- 1. Determine requirements essential to the development of improved techniques, equipment and facilities for conditioning, drying, storing and handling field crops.
- 2. Develop and test improved techniques and equipment for drying and storing field crops while maintaining desired standards of quality.
- 3. Develop and test improved techniques and equipment for the efficient handling of field crops yet minimize physical damage to the product.
- 4. Develop and test improved techniques and equipment to prepare field crops for market.
- 5. Establish design principles and improved facilities for commercial storage of field crops.

Progress - USDA and Cooperative Programs

A. Corn

1. Drying and storing. In cooperation with the Purdue Agricultural Experiment Station, the studies continued at Lafayette, Ind., showed that partial drying of 30 percent moisture field-shelled corn to 20 percent with heat and finishing the drying with aeration was only partially successful. Aeration airflow rates of up to 1/2 c.f.m. per bushel were not adequate to lower the corn moisture to safe levels in time to prevent substantial mold growth. Aeration was continuous. Late harvest, poor quality of the crop, and excessive fine material in the corn undoubtedly contributed to the poor results. Corn field shelled at 32 percent moisture and stored under aeration was maintained in satisfactory condition for only about a week. Corn dried in commercial models of concurrent flow dryers was of acceptable quality when dried with air temperatures up to 300° F. Above this temperature the bulk density and the milling score was reduced substantially. In corn metering tests using fluted rolls a more even flow was obtained at low corn moisture levels, at high flow rates, and with the rolls rotating clockwise in a 60° hopper.

2. Storing high-moisture corn. Under a research cooperative agreement with the Kansas Agricultural Experiment Station, studies were continued at Manhattan on the storage of high-moisture corn. Representative samples of the field-shelled corn used in the laboratory and refrigerated storage tests were analyzed for extent of mechanical and physical damage to kernels. Visual and screen separations and classifications were made on each initial sample. Unsound kernels due to small or large cracks in the seed coat and endosperm were classified as mechanical or harvester damaged. Broken kernels were hand separated. Small pieces and fine material were separated by a 12/64-inch prescribed sieve. October field-shelled test corn averaged 26.7 percent moisture, 0.9 percent fine material, 6.7 percent broken kernels plus 8.6 percent cracked seed coats. November field-shelled test corn averaged 26.5 percent moisture, 2.5 percent fine material, 11.2 percent broken kernels, plus 11.8 percent cracked seed coats. Fungal infestation and mold development were first apparent on mechanically damaged corn kernels and showed in seed coat cracks down into the endosperm. Corn samples taken on the prescribed days during the cooling and storage tests were found to yield three principal molds, Rhizopus, Penicillium and Mucor. Up to 30 percent of kernels were infected with Rhizopus in the 25 percent moisture corn at 35° F. but reduced as the corn dried. Penicillium increased at cold temperatures as long as the corn was high moisture and reached 60 percent near the center of the 6 ft. depth bins. Mucor continued to develop at low temperatures and ranged up to 78 percent after 100 days of storage. Test corn contained no visible nor vielded any toxin producing molds.

B. Wheat

1. Reducing damage from handling. In cooperation with the Kansas Agricultural Experiment Station, at Manhattan, significant effects of variety, method of cutting, location, and year on internal physical damage to wheat kernels were observed. Many internal cracks in kernels already were formed in the field before harvest. Only radial cracks due to wetting and drying cycle were observed. A mechanism of crack formation in kernels were postulated. average breaking strength of wheat kernels decreased linearly with increased internal damage. Frequency of breaking strength of kernels exhibited a normal distribution. An index for predicting or describing susceptibility to breakage of wheat kernels was defined. A linear relationship between index defined and percent internal damage was obtained. Mechanical damage of soybeans and corn at combines was examined. Breaking strengths of soybeans and corn kernels were evaluated. Significant effects of size, orientation, and moisture content of kernels on the breaking strength were observed. Studies on grain damage by the pneumatic conveying system were made with yellow corn samples to evaluate the effects of size and shape of kernels, air velocity, history of grain, and number of successive handlings on damage.

2. Aeration and drying. Also at Manhattan, Kans., studies were continued on the effectiveness of a two-fan, crossflow, ventilation system for conditioning high moisture, or tough, wheat. The two fans were controlled manually by the elevator operator or project personnel depending upon the test scheduled or seasonable weather conditions. The test bin at Abilene, Kans. was filled four different times with a total 1,000 bushels of new crop 1967 wheat. All four lots officially graded "tough." The first full bin was ventilated for two weeks starting July 1 for a total of 105 hours of fan operation. Averages from the moisture content sample test data indicated a reduction of 0.5 percentage points from 14.6 to the 14.1 percent. Recorded grain temperatures indicated cooling of 16.6° F. The second bin lot averaged 14.3 percent moisture, was held 10 days, ventilated for 135 hours resulting in moisture reduction of 0.2 percent and mean grain temperature reduction of 5° F. The third bin 1ot averaged 15.2 percent moisture, was held 16 days and ventilated for 232 hours. Representative grain temperatures were reduced 13° F.: the moisture content was reduced 1.4 percent to 13.8 percent. In August a fourth lot with moisture content averaging 13.7 percent was held for two weeks while the fans operated 135 hours. Average grain temperatures were reduced 10° F. and the final moisture content tested 12.8 percent. Tests of this one system over a 4-year period included 28 bins of grain, or more than 750,000 bushels, and the operation of the crossflow fans for over 3,000 hours.

Temperature variations of Hard Red Winter wheat stored in an 18-foot diameter bin with 100-foot grain depth without turning or aeration for 2 1/2-year period, have been analyzed. Variation in the vertical direction was practically negligible compared to that in the radial direction. Also changes in diurnal and annual ambient temperature can be represented as a periodic function of time. An analytical solution representing transient heat transfer within wheat stored in a concrete bir was derived from the Fourier heat condution equation. Temperature distributions in the wheat as generated by a derived solution agree well with corresponding actual temperature distributions. Differences in "lag" of temperatures between the ambient temperature distribution and grain temperature distributions increased almost linearly as the radial distance from the center of bin decreased. The analytical solution obtained should be useful in predicting grain temperature distribution, in effective scheduling of turning and mixing of grain, and in evaluating thermal diffusivity of grain for the case of transient heat transfer.

C. Sorghum Grain

1. Aeration. In cooperation with the Kansas Agricultural Experiment Station, a study of aeration of sorghum grain in commercial storages was continued at Manhattan. Analysis of data from tests were continued in an effort to explain the moisture loss factor for the grain and how it contributed to the overall weight loss or shrinkage. The important factors considered are (1) initial condition, grade, representative moisture content, mean temperature of grain; (2) location, seasonal weather conditions, time in storage; (3) size of storage units, upright or flat type storage and its structural material; (4)

type and design of aeration equipment and airflow rate; (5) handling, transfer and turning equipment used; (6) control and operating schedules for unit fans or systems. On an annual basis with comparable variables, grain held in steel flat storage buildings or small metal bins is exposed to more solar energy and higher temperatures than grain in upright storages, thus causing a greater moisture loss. Aeration fans should be operated at night for dry grain, but continuously for wet tough grain.

D. Rice

- 1. Drying. In cooperation with the Texas Agricultural Experiment Station, rice drying studies were continued at Beaumont. Pilot-scale drying tests indicated that the drying rate increased as heated air velocity was increased in the range between 115 and 167 cubic feet per minute with no significant difference in milling yield of rice. High air velocities resulted in the removal of a considerable amount of chaff and other trash from sound rice. Laboratory tests were conducted to study the effect of various rice temperatures and air velocities on drying rate and milling yield when rice was heated in vapor tight containers, then cooled by forced ventilation. Drying rate was increased at higher rice temperatures before cooling and was reduced by cooling in shorter times with high air velocities. There was no significant effect of either rice temperature or cooling air velocity on the milling yield of rice samples. Tests on mixing equal amounts of rice at two different moisture contents indicated that although 75 percent of moisture equalization occurs within the first 48 hours, equalization was not complete within a 40-day period. Undried rice storage tests showed that aeration using refrigerated air at 65° F. maintained rice at grade No. 1 for 10 days compared with six days for rice aerated with ambient air at 82° F. Storage mold, A. flavus and aflatoxins were inhibited when rice was aerated with refrigerated air.
- Handling rice at commercial dryers. Also at Beaumont, studies on handling rice during the drying operation at commercial dryers indicated that plant layout was a major factor affecting efficient operation. At some dryers changes in conveying equipment to allow simultaneous receiving, drying and shipping of rice were recommended. Partial loading of bucket elevators was a major bottleneck in rice handling operations. An ammeter at a remote location in the elevator motor circuit was effective to monitor loading. A photoelectric relay installed in an elevator boot to control the conveyor feeding the elevator aided in guarding against elevator choke-ups. scalper, or cleaner, commonly installed above a drying unit's garner bin, was a bottleneck when trashy rice was handled. A bypass was designed to alleviate this problem. Proper use of the dryer-aeration process reduced the required number of passes for drying lots of rice. Tests on handling milled rice by a bucket elevator indicated no physical damage resulted from conveying. Various belt speeds for elevator tested showed maximum handling capacity occurred at 250 feet per minute.

Grain

- 1. Effect of handling equipment. Under a research contract with Cargill. Inc., tests were continued to study the extent and causes of physical damage to grain by handling equipment used in commercial facilities. About 80 percent of the scheduled tests are complete. Corn was more subject to damage than wheat, soybeans or pea beans. Breakage resulting from a free drop of 100 feet was only 0.3 percent in wheat but ranged up to 13.8 percent in corn. Free drop of 100 feet caused 7-10 times more breakage in corn than caused by a bucket elevator or a grain thrower as they are normally used. In soybean tests, the breakage increased from 1.2 to 4.1 percent as free drop heights were increased from 40 to 100 feet. Dropping on concrete caused 50 percent more breakage than dropping on other soybeans. Dropping through a spout resulted in less breakage than free fall in an open stream. Also, there was less breakage in free fall from a 12-inch discharge than from an 8-inch discharge, a result which was unexpected. With bucket elevators, the damage increased with increased belt speed, but there was no difference between front and back feeding. Breakage was greater in soybeans at lower moisture levels and at lower temperatures.
- 2. Dynamic and static pressure phenomena in stored grain. Under a research contract with Dr. J. D. Isaacson, St. Louis, Mo., investigations continued on developing a basic theory of static and dynamic pressure phenomena in grain under storage conditions. With constant parameters except the varying wall-friction to study wall vibrations under dynamic conditions, the lateral pressure was 1.3 of normal static at midheight, and 1.6 at bottom of bin under normal friction conditions and nearly identical to pressures from low static friction conditions. Varying the ratio-function produced excessive dynamic lateral pressures in the upper two-thirds of the wall and a sharp increase in the lowest third. Study shows solving problems of grain pressures should be directed toward using numerical methods and digital computers.
- 3. Uniformly blending seed. Under a research cooperative agreement with the Mississippi Agricultural Experiment Station, the study was continued at State College on uniformly blending seed lots. The center-of-sidewall device was tested using layers of various colored plastic particles. Results showed three passes through the bin produced acceptable blending. Blending free-flowing seed having dissimilar physical characteristics (corn and grain sorghum) was tested with three of the devices. Five passes did not produce an acceptable blend. Separation occurred during free fall. Tests to blend a small amount of weed seed into sorghum showed three passes resulted in an average 26 weed seed per pound of sorghum rather than the theoretical 22.5.

4. Grain transport in boxcars. A study of defects and losses in 1989 boxcar shipments of grain was completed during the year. Of the boxcars inspected, 55 percent were classified as defective and the remaining 45 percent had no defects. Differences between recorded origin and destination weights for corn and soybean cars were so varied that definite conclusions about the relation between losses and defects could not be drawn. Analysis of wheat car data showed that cars with defective walls had a mean loss of 268 pounds more than cars without that defect. Cars with defective floors had a mean loss of 360 pounds more than cars without such defect. Cars with paper grain doors had a loss of 324 pounds more than cars with wooden doors. All of the differences were statistically significant.

The report on this work will be published as soon as it is cleared by the last USDA reviewing agency.

Publications - USDA and Cooperative Program

Corn

- Thompson, R. A. and Foster, G. H. 1967. Dryeration--high speed drying with delayed aeration cooling. ASAE Winter Meeting.
- McKenzie, B. A., Foster, G. H., Mayes, R. T. and Thompson, R. A. 1967. Dryeration--better corn quality with high speed drying. Cooperative Extension Service AE72. Purdue University.

Grain Sorghum

Hutchison, R. S. and Holman, L. E. 1968. Fan ventilation of air space above grain in flat storages. ARS 52-27.

Rice

- Louvier, F. J. and Calderwood, D. L. 1968. Drying and handling rice at commercial dryers. The Rice Journal.
- Louvier, F. J. 1968. Damage to milled rice during simulated bulk handling. The Rice Journal.
- Louvier, F. J. 1968. Studies of handling procedures at commercial rice dryers. Proceedings Rice Technical Group, New Orleans, La.
- Calderwood, D. L. 1968. Effect of varying air velocity in a continuous flow dryer. The Rice Journal.
- Calderwood, D. L. 1968. Storage of undried rice in aerated bins. The Rice Journal.
- Calderwood, D. L. and Schroeder, H. W. 1968. Aflatoxin development and grade of undried rice following prolonged storage in aerated bins. ARS 52-26.

COOPERATIVE MARKETING Farmer Cooperative Service

USDA and Cooperative Program

	: Scien	ntist Man-Yea:	rs F.Y. 1968
Location of Intramural Work	: RPA	:	Total
	•	•	
Washington, D.C.	: 2.8	:	2.8

Problems and Objectives

Cooperatives have become broader in scope and diversified in activities. No longer do most of them handle only one commodity or supply one service. Problems inherent in cooperative growth and difersification are organization, financing, and management. Companion problems include searching out new types of cooperatives and new services to meet the changing needs of rural America, improving cooperative structures so as to increase efficiency and inject a higher level of competition into the marketing system, and improving the economic power of farmers through cooperatives as a means of insuring equitable terms of sale for individual farmers.

Major objectives of this cooperative research are to develop and evaluate alternative ways to:

- 1. Achieve more efficient organizational and financial structures for more effective diversified operations.
- 2. Insure more effective member participation and control.
- 3. Improve effectiveness and identify opportunities for providing additional marketing functions.
- 4. Add, modify, or develop different service-type activities and identify product innovations that will help improve effectiveness.
- 5. Achieve for farmers, through their cooperatives, economic strength in bringing about terms of trade more favorable to them than those prevailing or that would prevail otherwise.

Progress - USDA and Cooperative Program

RPA 509 - MARKETING FIRM AND SYSTEM EFFICIENCY

A. Cooperative Sales, Services, Distribution, and Pricing Practices

Grain. Elevator design and size, type of equipment, and operating practices that will increase efficiency in cooperative grain marketing is under study in Ohio.

Regional grain cooperatives. Storage capacity for 23 regional grain co-ops has increased from 35.5 million bushels at the end of World War II to 298 million bushels by harvest time in 1968. Volume in 1966-67 was 980 million bushels, down from 1,030 million bushels the previous year but still the second highest on record. Net margins were off substantially -- the lowest since 1944-45.

B. Cooperative Trends and Potentials

Alfalfa dehydration. Work continues on a study to determine conditions necessary for establishing successful alfalfa dehydrating plants, evaluate the experience of cooperative and other dehydrating operations, and develop guidelines for successful dehydrating operation by cooperatives.

A study of 20 dehydrating plants (including both cooperative and proprietary) is nearing completion. In addition, records of a number of cooperative dehydrating plants that discontinued operations are being studied to determine causes of failure.

Preliminary observations indicate:

- 1. Successful alfalfa dehydrating operations depend heavily on a favorable location and prudent management.
- 2. Location, percentage of plant capacity used, and size of plant greatly influenced production costs.
- Cooperative and proprietary plants have had similar mortality rates.
- 4. Managers of cooperative alfalfa dehydrating plants must have considerable skill in supervising production and maintenance and repair, and have a good knowledge of markets.

Part of this work is being performed under contract by Montana State University.

C. Cooperative Organization, Finance, Management, and Member Relations

Grain. Two studies were completed, in cooperation with the University of Kentucky, to develop guidelines and recommendations for improving grain marketing facilities and practices in two areas of western Kentucky.

The study showed the need for seven new or expanded cooperative grain handling and storage facilities and several smaller grain loading stations to better serve grain producers. These new and expanded facilities should increase grain producers' income by about \$2 million annually.

A similar study is underway in a third area of western Kentucky.

The annual analysis of 23 regional grain cooperatives showed that these cooperatives continue to expand their services. Mergers and new facilities have paved the way for greatly increased efficiency.

Rice. A study conducted in cooperation with the land grant colleges of Arkansas, Louisiana, and Texas of the southern rice industry was completed. Major findings suggest that the industry should:

- 1. Cooperatively strengthen its market position through integrating processing and marketing functions.
- 2. Develop a strong unified sales organization that can efficiently export a large volume of rice.
- 3. Explore potentials for developing closer working arrangements between driers and mills that would result in better control over movement of rice from the driers to the mills.
- 4. Explore possibilities of bulk shipments of more rice in covered hopper cars to gain lower rail rates, even though transit privileges and other fringe benefits may be lost.
- 5. Commingle rice of similar quality to increase operating efficiency and improve use of storage facilities.
- 6. Consider integrating rice handling and marketing with one or more complementary crops such as soybeans.

Publications - USDA and Cooperative Program

- Thurston, S. K., Callahan, S. and Miller, G. F. 1968. The Need for Additional Grain Facilities in the Lower Green River Area of Kentucky. FCS Service Report 90.
- Thurston, S. K. and Callahan, S. 1968. Grain Facility Needs in the Pennyroyal Area of Kentucky. FCS Service Report 94.
- Traylor, H. D., Price, G., and Markeson, C. B. 1967. Costs of Drying and Storing Rough Rice in Louisiana and Texas. Marketing Research Report 799.

ECONOMICS OF MARKETING

Marketing Economics Research Division, ERS

USDA and Cooperative Program

	:		Sc	ientis	t Ma	an-Year	rs F	Y. 196	8	
Location of Intramural	:		I	Resear	ch]	Problem	n Are	ea	:	Total
Work	:	501	:	504	:	507	:	509	:	
	:		:		:		:		:	
Washington, D. C.	:	1.0	:	3.8	:	1.3		0.7	:	6.8
Arizona	•		:	0.4	:				:	0.4
California	:		:	0.6	:		:		:	0.6
Iowa			:	1.0	:		:		:	1.0
Mississippi			:	0.6	:		:		:	0.6
Oklahoma	:		:	1.0	:		•		:	1.0
Total	:	1.0	:	7.4	:	1.3	:	0.7	:	10.4

Problems and Objectives

Constant changes in new technology and innovations are occurring in the physical processes of marketing agricultural commodities. These changes lead to reduced marketing costs with subsequent impacts on incomes to farmers, the competitive position of input and output marketing firms, and prices to consumers. The role of economic research is to provide more precise measures of these impacts and to analyze their effects on the performance of the marketing system.

As a greater share of our agricultural production goes into processing, the gap between what the farmer receives and what the consumer pays widens. In view of this, the need for improved forecasts of supply, demand, and prices of farm products are essential to more efficient and orderly marketing. Individual producers, processing and marketing firms, Government policymakers, all require objective information on future supply, demand and price conditions to make sound decisions. This type of information is of special importance to small firms, farmers, and consumers who do not have the resources for analyzing the many interrelated factors that must be considered in forecasting supply, demand, and price.

Competitive relationships in agriculture change constantly with the development of new technology, shifts in costs and consumer tastes, and organizational changes in the production, marketing, and farm supply sectors. Information on the nature, extent, and ramifications of these changes provide a firmer basis for public policy and private investment decisions.

More than \$100 billion of consumer expenditures for agricultural products go for marketing, processing, and transportation services. The potential returns to improve efficiency are large but the adjustments within individual sectors must be coordinated with other sectors of the system to optimize results. In addition, improved measures of the performance of the marketing system and its subsectors are vital if these potential savings are to be realized.

Major objectives of this research are to:

- 1. Measure farm-to-retail price spreads and their components.
- 2. Determine alternative methods that result in increased efficiency.
- 3. Analyze the optimum size, number, type, and location of marketing facilities.
- 4. Develop market system specifications required to provide efficient services at minimum cost.

- 5. Measure the effects of changes in supply of individual commodities upon farm prices, marketing spreads, and consumer prices.
- 6. Determine and analyze major factors affecting locational advantages of industries engaged in the marketing, processing, and distribution of agricultural products.
- 7. Measure the impact that changes in major locational factors and Government programs have on regional competition and resource allocation.

Progress - USDA and Cooperative Programs

RPA 504 - PHYSICAL AND ECONOMIC EFFICIENCY
IN MARKETING FIELD CROPS

A. Marketing Margins for Bread

Bread prices have continued their upward trend almost uninterrupted since 1947-49. The U.S. average price in 1967 was 22.2 cents per pound loaf sold in food stores. But the rate of increase per year has slowed down to 1.8 percent during the 1960's from 3.2 percent during the 1950's. Most of the increase has occurred in the baking segment, followed by retailing. The farm value of ingredients used in a pound of bread was 3.4 cents in 1967, only 0.1 cent above the 1947-49 average, while the retail price of bread in the first two quarters of 1968 has averaged just slightly below the 1967 average.

B. Grain Storage and Handling Cost

Research relating to the cost of storing and handling of grains in public storage facilities will be continued and updated. A survey of 105 of the participants included in the 1965 original survey will be conducted to determine the nature and extent of changes that may have occurred since the base period. This research provides a basis for many public policy decisions on grain storage and is a vital input for our analyses of the grain marketing system.

C. Rice Storage and Handling Cost

A study of the cost of drying, storing and handling, and milling rice was initiated. Detailed cost estimates have been obtained from a sample of 32 firms from three major producing areas (Arkansas-Mississippi, Louisiana-Texas, and California). This detailed information will enable

costs to be developed for each major function of these operations. These functional cost data and analyses will enable management to appraise the efficiency of their operations and to see the effect of such factors as size, volume handled, and location on costs. These data also will provide information needed in the development of an overall system analysis of the rice industry.

D. Transportation and Location Economics

A spatial equilibrium model of the domestic grain industry has been developed. Preliminary results indicate a considerable suboptimization of the storage-processing-distribution system. The influence of alternative storage programs on the transportation system and the effect of alternative transportation rates on the grain marketing system are being studied using this model. The model is being updated and refined.

Incorporating previously developed estimates of costs for handling and storage and for transportation, the feasibility of extending the 9-foot Missouri River barge channel from Sioux City, Iowa to Yankton, South Dakota was studied at the request of the Army Corps of Engineers. A minimum cost flow analysis indicated that such an extension of the channel is feasible and would increase the flow of grains and oilseeds down the Missouri from the study area by some 760 thousand tons.

The effect of changes in transportation rates on the location of the flour milling industry is receiving special attention and a report will be completed this year identifying the location of excess milling capacity and the economic costs of alleviating it via use of alternative transportation rate structures. This work is being conducted in cooperation with Iowa State University and is a vital part of the refinement of analyses of the grain marketing system. It also relates directly to the objectives of the Mid-America Governors' Transportation Committee.

The archaic distribution system and redundant services performed by whole-sale bakeries account for a large proportion of the cost of bread. A model regional organization and distribution system for the wholesale bakery industry is being developed in cooperation with the University of California and the local industry. Preliminary results confirm substantial inefficiencies in distribution methods and practices. Results from studies such as this, when combined with those of other sectors of the industry, provide the refinements necessary for evaluating the entire grain and grain products marketing system and developing suggestions for improvement.

RPA 507 - COMPETITIVE INTERRELATIONSHIPS IN AGRICULTURE

A. Grain

A study of the structure and practices of the grain marketing industry in the Western States was completed that complements similar and completed research for the other grain producing regions. There was no significant trend towards integration among country elevators, flour mills or feed manufacturers in the region. The means utilized by firms within these industries to transport grain and grain products was quite stable over the 4-year study period.

RPA 509 - MARKETING FIRM AND SYSTEM EFFICIENCY

A. Feed Grains

In order to assist in production planning of feed manufacturers, a composite formula model was developed and is being used commercially for least-costing single feed formulations. A production planning model which will least-cost feed formulations simultaneously taking into account operation restrictions and inventory has also been developed and is being tested by a cooperating feed manufacturer. The latter model is performing well in planning ingredient procurement two and three weeks in advance. These models draw heavily on previous costs analyses of feed manufacturing and are designed to provide management with a total systems approach to profit planning and cost control.

Publications - USDA and Cooperative Program

- Coffin, H. G. and Reilly, W. R., September 1968. Rail Freight Rates: Potential Reduction on Corn Shipped to New England. University of Connecticut, Storrs Agr. Exp. Sta. Bul. 407.
- Eiland, J. C., July 1968. Marketing Spreads for White Bread. Misc. Publ. 1091, 15 pp.
- Taylor, Reed D., Kohler, George O., Maddy, Kenneth H., and Enochian, Robert V., January 1968. Alfalfa Meal in Poultry Feeds--An Economic Evaluation Using Parametric Linear Programming. AER-130, 19 pp.
- Vosloh, Carl J., Jr., March 1968. Costs and Economies of Scale in Feed Manufacturing. MRR-815, 66 pp.

CONSUMER ATTITUDES AND PREFERENCES Standards and Research Division, SRS

USDA and Cooperative Program

	:_	Scientist	Man-Years	F.Y. 1968	
Location of Intramural Work	:	RPA 508	:	Total	
	:		:		
Washington, D.C.	:	0.3	:	0.3	

Intramural program is supplemented by extramural support representing 0.5 SMY's at private market research firms.

Problem and Objectives

Domestic consumption of agricultural commodities depends on the behavior of some 200 million consumers. But, in our complex marketing economy, it has become almost impossible for consumers to discuss their opinions, preferences, and complaints with producers and marketers. Knowledge of consumer reactions to agricultural products is becoming increasingly important because we are in a period of rapid change: There is a growing challenge to farm products and farm income from a wide variety of competitive products of nonagricultural origin; there is a proliferation of mixtures, forms, processes, blends, and other innovations affecting farm products; there is an increasing awareness of the adverse effects incorrect decisions made as a result of inadequate information on the consumer's viewpoint can have on USDA functions as well as farm income.

Major objectives of this market development research are to provide:

- 1. A line of communication from consumers back to those concerned with production, processing, and marketing farm products.
- Identification of areas on which technical research should focus
 to provide farm products with characteristics that will increase
 consumer acceptance.
- 3. Evaluation of consumers' reactions to specific product variations under controlled laboratory conditions.
- 4. Assessment of consumers' knowledge and opinions concerning
 Department functions such as educational programs, setting grades
 and standards, and inspection of farm products.

Progress - USDA and Cooperative Program

RPA 508 - DEVELOPMENT OF DOMESTIC MARKETS
FOR FARM PRODUCTS

Survey of Homemakers Use of Potatoes, Rice and Wheat

The final report is being prepared for publication for a nationwide study among homemakers on their use of and opinions about selected potato, rice, and wheat products. Almost all of the homemakers interviewed reported that they had used white bread and some form of white potatoes and rice during the year preceding the interview. In general, the homemakers felt that fresh white potatoes, regular uncooked rice, and bread were high in food value, fattening, and low cost per serving.

Among respondents who said they had used some processed potato or rice products, the feeling for the most part was that these products were higher in cost per serving than if prepared "from scratch" by the homemakers, but they were used primarily because they were easier or quicker to prepare.

Publications - USDA and Cooperative Program

None

SUPPLY, DEMAND AND PRICE ANALYSES (RPA 506)

Economic and Statistical Analysis Division, ERS

USDA and Cooperative Program

	:		:	Scientist
Location of Intramural Work	:	Commodity	•	Man-years FY 1968
			:	
Washington, D. C.	9	Food Grains	:	2.0
	•		:	
Washington, D. C.	•	Feed	:	2.0
	:		:	
		Total	:	4.0

Problems and Objectives

Farmers, processors, distributors, policy officials, and others need continuing economic intelligence regarding supply, demand, and price prospects to aid them in making sound production and marketing decisions to help assure adequate, well-balanced supplies of food and fiber. To insure that the situation and outlook work is as accurate and precise as possible, continuing research is needed on supply response to price, effect of supplies on price, and the effect of changing demand on prices and income.

Specific Objectives:

- 1. Provide research needed for strengthening situation and outlook work.
- 2. Improve forecasts of future supply and demand prospects.
- 3. Appraise market situations and prospects to aid decision makers in making sound and profitable decisions.
- 4. Appraise current and proposed programs to aid officials in formulating agricultural programs and policies.
- 5. Develop and improve basic statistics for use in statistical and economic analysis.

Progress - USDA and Cooperative Programs

A. Food Grains

Emphasis in 1967 - 1968 was directed to the problem of declining wheat prices in the face of record crops and rising carryovers; and to appraisals of suggested policy and program alternatives for improving prices. Two special problem areas were the record large soft red winter wheat supply and the poor export potential for all wheat. As a result, work was directed toward estimating (1) the probable expansion in use of soft red wheat as feed and (2) its substitution for other classes in the milling industry. Attention was placed on the feasibility of substituting soft wheat for other classes in exports to countries receiving food aid.

With lower wheat prices and renewed interest in the price support loan program, studies were made to forecast price levels and loan activity from data available early in the marketing year. Work performed to date indicates that (1) the season average farm price in 1968/69 may be only slightly above the national average loan, and (2) the quantity of 1968 crop wheat placed under loan will be the largest since 1958, somewhat more than 400 million bushels. Other analytical work has centered around the relationship of wheat stocks in the major exporting countries and the price of one of the major wheats in world trade. It is planned to broaden this to cover a

number of wheats in world trade, particularly U.S. wheats and foreign competitors. Research to determine the factors associated with wheat feeding, on a wheat and feed-grain region basis, has shown only limited usefulness to date.

Study of a new price supporting mechanism, a commercial warehouse reseal loan program, was carried out to assess its effectiveness and acceptability. Analyses of various alternative wheat allotment and diversion programs for 1969 were also conducted, the objective of which would be to bring supplies into better balance with demand and improve prices to producers.

Other special studies were made on (1) the export payment rate for durum wheat, (2) probable spring wheat production taking into account a new variety of wheat, (3) wheat production and income in the absence of a price support and allotment program and (4) evaluation of the International Grains Arrangement.

B. Feed

Two important changes have occurred in the feed grain picture in recent years. During 1960-66, supplies and Government stocks were reduced from burdensome levels, and prices increased. In 1967 and 1968 bumper crops increased supplies to relatively high levels, bringing lower prices to producers.

Favorable livestock/feed price relationships in 1968/69, however, are expected to stimulate increased domestic use, bringing supply and disappearance into closer balance than in 1967/68.

An analysis on trends and variations in corn yields over the past 50 years revealed: (1) No upward trend in yields during 1916-35, (2) moderate upward trend during 1935-51 and (3) a pronounced upward trend during 1951-65. Factors contributing to changes in the yield trends in these 3 periods included: (1) Unfavorable weather and low prices in the depression years, (2) hybrid seed, acreage reduction in the lower yielding areas and favorable prices during and following W.W. II, and (3) expanded use of fertilizer, increased plant population, weed and rootworm control and irrigation in western areas.

Hay production, pasture-feed conditions, number of roughage-consuming live-stock and prices received by farmers were the most important factors associated with changes in hay prices during the past decade. Three estimating equations were developed from various combinations of these factors which statistically explained 85 to 95 percent of the change in price. The results indicate that a 10 percent increase in hay production or pasture-feed condition would be followed by a decline of about 10 percent in price. A 10 percent increase in roughage-consuming livestock increases the price by 10 to 12 percent. And a 10 percent increase in farm prices of beef cattle and dairy products brings a 5 percent increase in the hay price.

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